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MYCOTAXON

AN INTERNATIONAL JOURNAL FOR RESEARCH ON TAXONOMY & NOMENCLATURE OF FUNGI, INCLUDING LICHENS

Volume LXX January-March 1999

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HOHENBUEHELIA ILERDENSIS, A NEW GRAMINICOLOUS SPECIES FROM SEMISTEPPE AREAS OF LLEIDA (WESTERN CATALONIA, SPAIN)

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KEY WORDS: *Hohenbuehelia*, chorology, Catalonia, Spain.

SUMMARY. *Hohenbuehelia ilerdensis* Courtec., Vila et Rocabruna, a new species growing at the bases of dead grasses in semisteppe communities near Lleida (Catalonia, NE Spain), is described. It is readily distinguished from related species such as *H. culmicola* Bon and *H. longipes* (Boud.) Moser by its small size, glabrous cap covering, typically central stipe, more or less violaceous colour, inconspicuous pleurocystidia and the presence of abundant caulocystidia.

RESUMEN. Se describe una nueva especie recogida en la base de gramíneas, en comunidades semiesteparias próximas a Lérida, caracterizada por su estípite típicamente central, su glabra cutícula, su pequeño tamaño, su coloración más o menos morada, la poca diferenciación de los pleurocistidios y la presencia de abundantes caulocistidios, lo que la separa bien de las especies más próximas, *H. culmicola* Bon y *H. longipes* (Boud.) Moser.

INTRODUCTION

During our investigation of the fungal flora that characterizes the dry plains and hills of the Ebro depression in Catalonia, we made numerous collecting trips in shrub vegetation corresponding to various successional stages of the local climax, the macchia of *Rhamnus lycioides* and *Quercus coccifera*. On clay-rich and more or less saline soils, in a community of *Artemisia herba-alba* and *Salsola vermiculata*, resulting from a partial recolonization of abandoned almond and olive orchards, we have found a rich community of saprotrophic fungi, producing ephemeral sporophores after rather heavy rains. Rain is very irregular in this area, amouting to a mean annual rainfall of about 400 mm. One interesting component of the Mycota is a small agaric growing abundantly between dead grasses remnants. A long period of intensive collecting yielded only two collections in the same area. We concluded that it is a new species of *Hohenbuehelia*, which we describe here.

HOHENBUEHELIA ILERDENSIS R. Courtec., Vila et Rocabruna *sp. nov.*

Fig. 1-2

ETYMOLOGY. From Ilerda, the latin name of Lleida (in Catalan) or Lérida (in Spanish), the town located 15 km east of the collection point in the Central Depression of Catalonia.

Pileus (7-)15-20 mm in diametro, convexus, deinde applanatus vel ad maturitatem depressus, glabratus, laevis, nitidus, violaceo- vel rubro-brunneus. Margo non striatus. Lamellae decurrentes, crassae, albae, dein leviter pallide brunneo maculatae. Sporae albae in cumulo. Stipes (12-)14-20 x 2-3 mm, centralis vel subexcentricus, cylindraceus vel curvatus, griseus, tomentosus, dein glabrescens. Carne albida, basim stipitis versus pulchre obscurior. Odore et sapore farinosi. Sporae (7-)7.5-10(-11) x (4-)4.5-5.5(-6) µm (Q = 1,5-2,2) allantoideae vel cylindricae, hyalinae, inamyloideae, laeves. Basidia 28-40 x 4-9 µm, elongatae, cylindrico-clavatae, tetrasporibus. Trama hymenophoralis subregularis vel sinuosa, ex hyphis 3-10 µm in diam., hyalinis, parietibus pulchre incrassatis et refringentibus. Pleurocystidia adsunt sed rara, submarginalia et faciliter collapsata, 20-40(-50) x 4-12(-14) µm, fusiformia vel cylindrico-clavata, parietibus tenuibus, circa 1 µm crassis, metuloidibus, cristallibus spectabilibus, usque ad 30 µm in diam. coronatis. Cheilocystidia numerosa, (20-)30-40 x 4-7(-8) x 1-2 µm, lageniformia, ad basim ventricosa et ad apicem tenua, simplicia vel bifida usque ad multifida vel difforme-nodulosa, cum rara vel nulla corpus glutinosus. Caulocutis ex hyphis erectis, attenuatis vel obtuso-capitatis, cheilocystidiis similibus, circa 10-18 x 2-2.5 µm, corpus glutinosus adest. Suprapellis (ixocutis) usque ad 250 µm crassus, ex hyphis 2-5(-6) µm in diam., repentibus vel pulchre erectis, obtusis, curvatas vel ad apicem leviter incrassatis (5-7 µm in diam.). Pigmentum parietale incrustatum. Fibulae copiosae.

Holotypus prope vico Castelldans dicto, in Catalonia occidentalis (Hispania), a J. Vila et X. Llimona lectus, 21-11-1994, 360 m altitudinis, ad Brachypodio retuso basim, in herb. LIP (Herbier de la Faculté des Sciences Pharmaceutiques et Biologiques de Lille, France) *sub nº* SP/94001 *conservatur. Isotypus in BCC-SCM n°3234. Topotypus (17.12.1996) in BCC-SCM n°3235*

Macroscopic description (Fig. 1):

Pileus (7-)15-17-20 mm in diam., convex when young, later flattened to slightly depressed in age. Cuticle glabrous, smooth, glossy, violaceous to reddish brown, with fine, concolorous, scattered granulations in some of the young basidiomes; margin not striate nor incurved. **Gills** decurrent, thick, at first white when young, later sometimes spotted light brown. Edge white. Lamellulae abundant. **Spore-print** white. **Stipe** (12-)14-20 x 2-3 mm, usually central, but in some cases slightly excentric, cylindrical, sometimes bent, slightly thicker in the apical part, greyish from the abundant tomentum at first, then gradually darker as this tomentum disappears. **Flesh** thin, white, slightly darker near the base of stipe; smell and taste farinaceous.

Microscopic description (Fig. 2):

Spores (Fig. 2a) (7-)7.5-10(-11) x (4-)4.5-5.5(-6) µm (Q = L/l variable: 1,5-2,2), allantoid or phaseoliform to cylindrical, rarely ellipsoidal, the inner side often concave and sometimes slightly angular, hyaline, smooth, inamyloid, with a large oil-drop or numerous fine droplets. **Basidia** (Fig. 2b) 28-40 x 4-9 µm, elongate, clavate-cylindric, 4-spored, clamped.

Sub-hymenial layer dense, relatively thin (up to 10-15 µm), with slender, tortuose, densely interwoven hyphae forming a structure looking like small puzzle pieces. **Gill trama** interwoven to subparallel, made up of hyphae 3-10 µm broad, hyaline but with slightly thickened (up to 0.5 µm) and glassy walls, more or less anastomosing or branched. **Metuloid pleurocystidia** (Fig. 2c) present (and often located close to the gill edge) but very easily overlooked because they are uncommon and heavily covered and hidden by crystals (furthermore, when the crystals become detached or get

dissolved, the cystidia seem to collapse easily) : 20-40(-50) x 4-12(-14) µm, fusiform to clavate-cylindric, with relatively thin walls, about 1 µm, bearing an enormous cap of very coarse crystals, up to 30 µm in diam. These pleurocystidia may differentiate laterally (not terminally) from the sub-hymenial hyphae. **Cheilocystidia** (Fig. 2d) very numerous (gill edge sterile or almost so), 20-30(-40) x 4-7(-8) x 1-2 µm, lageniform with an inflated base and a thin, sometimes irregular, bifurcate or even multi-furcate, rarely nodulose apex, the apex (or apices) often slightly inflated. Between these typical cheilocystidia are mixed a very few pleurocystidioid cells (metuloids identical to the pleurocystidia). Cystidia with hour-glass-shaped apex surrounded by mucoid droplet not seen on the gill edge. **Caulocutis** with numerous right-angled and erect cheilocystidia-like end-cells (Fig. 2e), tapering or slightly capitate, about 10-18 x 2-2.5 µm. At their tips may be found mucoid droplets, 2-5 µm in diam., rather difficult to observe because of the abundance of superficial crystals. **Stipe trama** wavy, with slightly interwoven hyphae; sometimes slightly diverticulate and with a parietal pigment. **Suprapellis** a rather thick ixocutis (about 250 µm) made up of hyphae 2-5(-6) µm broad, repent but with some erect, obtuse, curved or straight and slightly inflated tips (5-7 µm broad) (Fig. 2f). **Pigment** parietal, encrusting and strongly striped but apparently developing a more punctate or sandy-punctate and rather unusual pattern in age. **Clamps** abundant in all part of the basidioma.
MATERIAL EXAMINED. LLEIDA: Castelldans (Les Garrigues), UTM 3154596, alt. 360 m, on dead bases of the grass *Brachypodium retusum*, 21-11-1994, leg. J. Vila and X. Llimona, nº herbarium SP/94001 in herb. Courtecuisse (LIP) (*Holotypus*) and n° 3234 in BCC-SCM (*Isotypus*). Same locality, same collectors, 17-12-1996, n° SP/96001 in herb. Courtecuisse (LIP) and n°3235 in BCC-SCM (*Topotypus*).

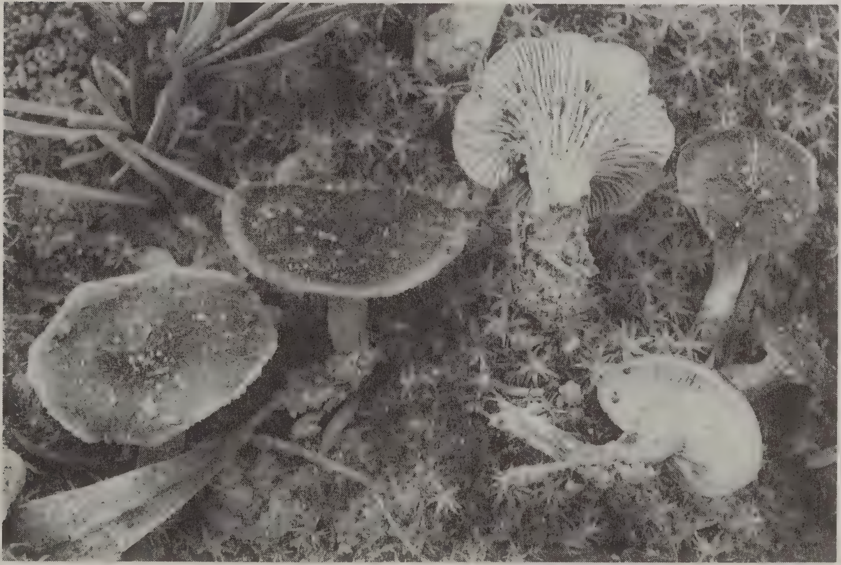

Fig. 1: *Hohenbuehelia ilerdensis* Courtec., Vila & Rocabruna. Basidiomes in their natural habitat.

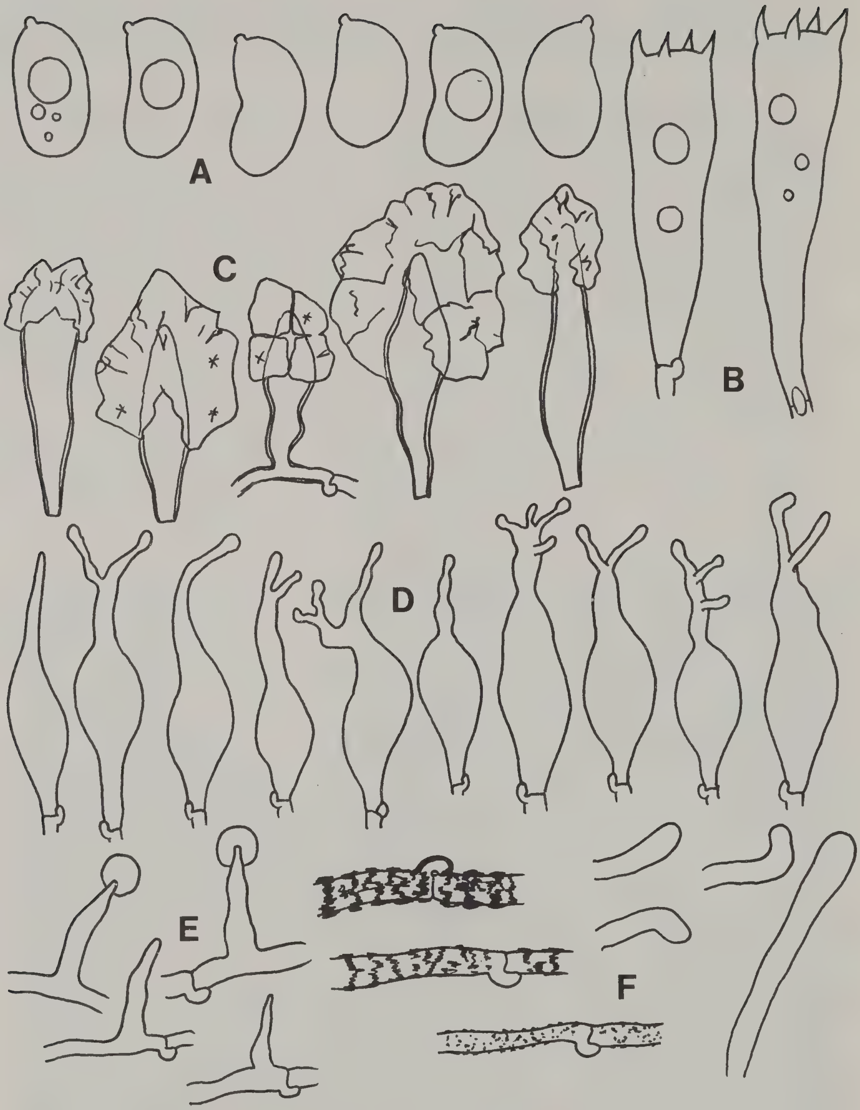

Fig. 2: *Hohenbuehelia ilerdensis* Courtec., Vila & Rocabruna. Microscopical features. A: Spores; B: Basidia; C: Metuloid cystidia; D: Cheilocystidia; E: Mucoid droplets on hyphae from the stipe covering layer; F: Some hyphae of the pileus covering.

DISCUSSION

The glabrous cuticle, the violaceous tinge, the atypical pleurocystidia, the almost always central, grey stipe, with abundant caulocystidia and the ecological preference for the basis of dead grasses (to date only *Brachypodium retusum*) are the main characters of this new species. *H. culmicola* Bon, growing on *Ammophila arenaria* in coastal dunes, has a densely tomentose, brownish-grey to sepia brown pileus, a lateral or excentric stipe, the flesh devoid of any distinctive smell or taste, typical and bigger (40-75 x 6-12.5 µm) pileocystidia and pleurocystidia. Another related species, *H. longipes* (Boud.) Moser, growing in *Sphagnum* mats in alpine or boreal areas, shares the farinaceous smell and the central stipe with our species, but the latter is longer [up to 40-60-(-80) x 3-5-(-8) mm], isabelline-coloured, devoid of caulocystidia, its pileus is brown-ochraceous to isabelline, 30-60 mm in diam., its basidia are usually bisporic (Thorn and Barron, 1986), or bi- to tetrasporic (Watling and Gregory, 1989), and it has larger pleurocystidia (75-100 x 8.5-14 µm).

ECOLOGY

Two collections have been found at the same spot, near Castelldans (Les Garrigues), at an elevation of 360 m, in an abandoned field colonized mainly by grasses (*Brachypodium retusum*, *Stipa capillata*) and *Artemisia herba-alba*. This locality is in the potential area of the *Rhamno lycioides-Quercetum cocciferae* Br.-Bl. et O. Bolòs (1954) 1957. Other fungi collected close to this point were: *Agrocybe vervacti* (Fr.:Fr.) Sing., *Coprinus spilosporus* Romagn., *C. vosoustii* Pilát, *Entoloma rusticoides* (Gill.) Noord., *Hohenbuehelia chevallieri* (Pat.) Pegler, *Lachnella alboviolascens* (Alb. & Schw.:Fr.) Fr., *Marasmius corbariensis* (Roum.) Sing., *M. quercophilus* Pouzar, *Melanoleuca leucophylloides* (M. Bon) M. Bon, *Montagnea arenaria* (DC.) Zeller, *Omphalina obscurata* Kühn. ex Reid, *Omphalotus olearius* (DC:Fr.) Sing., *Ramicola iberica* (Moreno & Esteve-Rav.) M. Bon, *Tulostoma brumale* Pers.:Pers., *T. kotlabae* Pouzar, *T. nanum* (Pat.) Wright and *T. xerophilum* Long.

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THREE SPECIES OF MICROTHYRIUM FROM NOLINA

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Abstract: Three new *Microthyrium* species are described, all from dead leaves of species of *Nolina*. Ascospores of *M. nolinae* and *M. guadalupensis* have novel placement and numbers of appendages. Ascospores of *M. arizoniensis* lack appendages. *Zalerion guadalupensis*, the anamorph of *M. guadalupensis* is also described.

Key words: appendaged ascospores, ascomycete, *Microthyrium, Nolina,* systematics, *Zalerion.*

INTRODUCTION

The genus *Microthyrium* Desmaz. has superficial shield-shaped ascomata containing bitunicate asci with their bases anchored at the inner periphery of the ascomatal wall and apices oriented toward the ostiole. The asci are interspersed with paraphysoids which may or may not deliquesce prior to ascospore release. Ascospores are 1-septate, hyaline, and may or may not bear appendages. More than 150 species, mostly tropical, have been descibed. More than 20 species have been removed to other genera or their placement in *Microthyrium* has been questioned, but the majority remain undisturbed formally. Recent estimates of the number of species are 10 (von Arx and Müller 1975) or 50 (Hawksworth *et al* 1995). A few European species have been considered synonymous. Müller and von Arx (1962) placed *Microthyrium cytisi* Fuckel, *M. moravicum* Petr., *M. pinophyllum* (Höhn.) Petr., *M. platani* Richon *apud* Sacc., *M. quercus* Fuckel, and *M. xylogenum* Bomm., Rouss., & Sacc. into synonymy with *M. microscopicum* Desmaz., the type species. Detailed studies (Ellis 1976) showed at least *M. cytisi* and *M. pinophyllum* are distinguished from *M. microscopicum* by absence, or presence and distribution of ascospore appendages and structural details of the upper ascomatal wall. Appendages were reported on young *M. pulchellum* Speg. ascospores in the 19th century, and finally, in the 20th century, noted for *M. macrosporum* (Sacc.) Höhn. (Müller and von Arx 1962). Unstained appendages are usually not visible, so the presence and possible value of the character was not recognized for more than a century after the first species was described. Appendages are now known for several species, but the genus as a whole has not been surveyed for their presence.

Nolina Michx. is found in the arid southwest of North America. The genus is now usually placed in the Agavaceae. Like other members of that family, it has a large woody caudex which may be wholly subterranean or form a distinct trunk. The leaves are narrowly linear, sometimes quite rigid, and remain attached to the caudex by a broad base for several years while decomposition proceeds. No fungus resembling *Microthyrium* has been recorded for this genus.

MATERIALS AND METHODS

Despite several trials, no growth resulted when free ascospores in a drop of sterile tap water were spread with a glass rod over the surface of commercial potato dextrose agar (PDA, Difco) in petri plates. Cultures were therefore prepared by dissecting ascomata, separating groups of asci, and washing them three times in sterile tap water. Groups of asci were then placed on PDA in a petri plate and samples were later isolated onto PDA slants in test tubes. Cultures were maintained at 23-25 C. Growth originating from groups of asci may be suspect as contamination, but the same results were obtained repeatedly, and colonies from the three species being studied were similar, not identical, so growth probably did not originate from contaminants present on or among the groups of asci isolated. Ascospore appendages were stained with lactophenol-cotton blue in early preparations, but it was found that the same stain without phenol gave better results as Ellis has suggested (1976).

TAXONOMY

Microthyrium nolinae A. W. Ramaley, *sp. nov.*

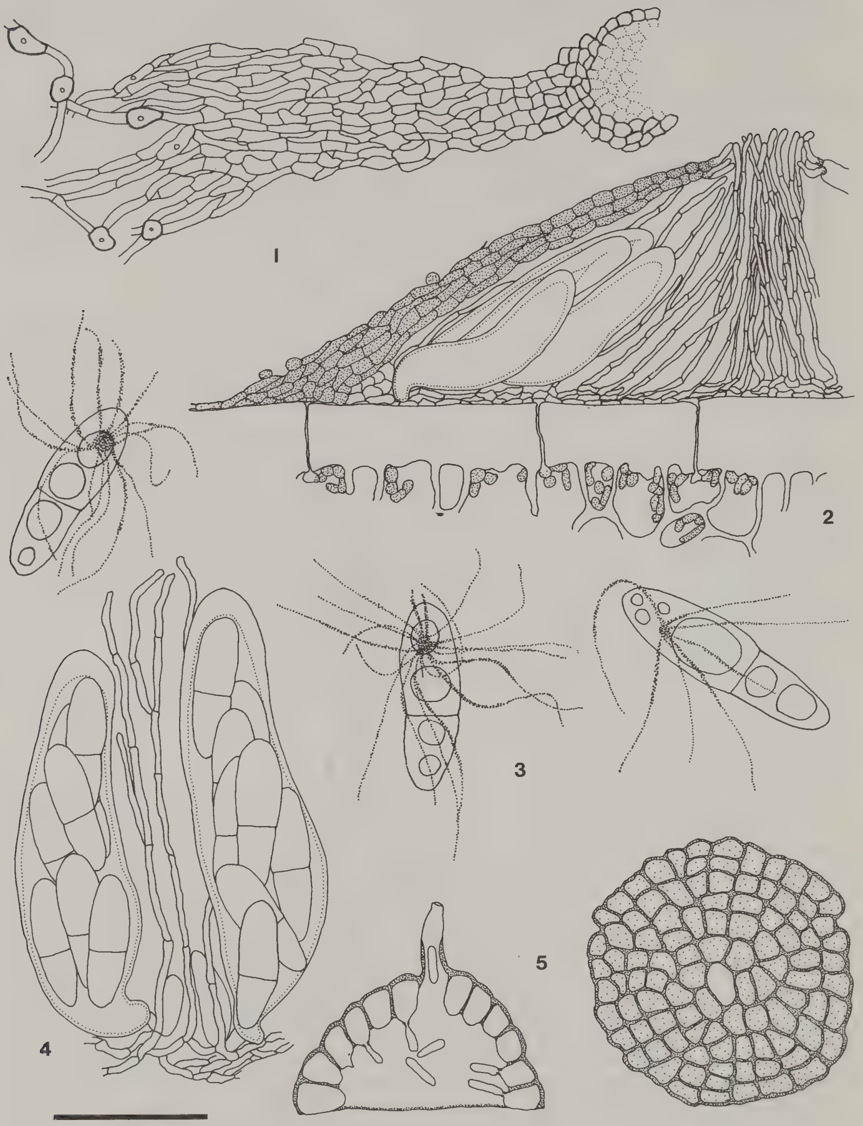
Mycelium superficiale inconspicuum, hyphopodiis, mycelium internum in cellulis epidermalibus, parenchymatibus, fibris. Ascomata superficialia, prope basin foliorum emortuorum, scutata, 150-400 μm diam; supra peridium ca 10-15 μm crassum, ex 3 vel 4 stratis cellularum parvarum, brunnearum, aliquantum crassitunicatarum, complanatarum constans, stratum basale tenue, cellulae hyalinae, tenuitunicatae. Hamathecium filis ramosis, anguste cellulosis inter ascos et columnam centralem formans. Asci numerosi, bitunicati, oblongi vel obclavati vel aliquantum ventricosi, leviter inequilaterales, 48-72 x 14.4-16 μm, ascosporis 8 irregulariter dispositis. Ascosporae obovoideae, 16-21.6 x 5.2-6.4 μm, 1-septatae submedianae vel medianae, hyalinae, tenuitunicatae, plerumque guttulae 2 magnae utraque cellula, tunica hyalina carentes, usque ad 20 appendiculis delicatis usque ad 18 μm longis affixis a latere ad nodum in hemispora supera ca 1/3 distantia ab apice ad septum. Ascomata coelomycete scutato concomitata. Conidiomata superficiales, unilocularia, 15-40 μm diam, paries superus ex strato 1 cellularum brunnetunicatarum radiantium, paries internus nullus, basis ex pellicula brunnea constans. Ostiolum ex tunica cellula tubum formanti usque ad 8 μm elatum constans, circa in medio *conidiomatis. Cellulae conidiogenae phialidicae, determinatae, discretae, hyalinae, leves, paries formantes. Conidia bacillaria, 2.4-3.2 x 0.5-0.8 μm, aseptate, hyalina, tenuitunicata, levia.*

HOLOTYUS: In foliis emortuis Nolina micrantha I. M. Johnst., "U. S. A. TEXAS: Brewster County, S. of Fort Stockton, U. S. Hwy 385, mile 511.5, roadside plants, 20 May 1995," *a Annette Ramaley n. 9524 lectus in BPI depositus. Paratypus in foliis emortuis Nolina micrantha* I. M. Johnst., "U. S. A. NEW MEXICO: Lincoln County, Valley of Fires, U. S. Hwy 380, mile 61.15, 24 October 1996," *a Annette Ramaley n. 9611 lectus in UC depositus.*

Superficial mycelium inconspicuous, forming hyphopodia (FIG. 1), internal mycelium in epidermal, parenchyma, and fiber cells, connected to the ascomata and superficial mycelium by thin hyphae (<0.5 μm) through the epidermal cell wall and cuticle to hyphopodia or occasionally more normal appearing mycelial cells or apparently morphologically undifferentiated cells of the basal layer of ascomata. Ascomata (FIG. 2) superficial near the base of dead leaves, scutate, outline approximately circular to somewhat irregular, occasionally confluent, 150-400 μm diam, dark, apex rounded, ostiole 20-30 μm diam at maturity; upper peridium ca 10-15 μm thick, composed of 3 or 4 layers of small, brown, somewhat thick-walled, flattened cells, basal layer thin, the cells hyaline, thin-walled; peridium surface of approximately isodiametric cells near the ostiole, becoming rectangular away from the ostiole (FIG. 1). Hamathecium of branched, narrowly cellular filaments between the asci and forming a central column (FIG. 2, 4), persisting while spores are shed, forming a central plug in the ostiole. Asci (FIG. 4) numerous (>30 in mid-size ascoma), maturing successively, obliquely directed toward the pore, bitunicate, oblong to obclavate to somewhat ventricose, slightly inequilateral, 48-72 x 14.4-16 μm, with 8 irregularly arranged ascospores. Ascospores (FIG. 3) obovoid, 16-21.6 x 5.2-6.4 μm, submedianly or medianly 1-septate, septum 0.50-0.62 distance from apex to base, hyaline, thin-walled, each cell usually with 2 large guttules, lacking a hyaline coating, up to 20 delicate appendages (FIG. 3) up to 18 μm long attached laterally on the upper hemispore about 1/3 the distance from apex to septum. Ascomata accompanied by a scutate coelomycete. Mycelium that of the ascomata. Conidiomata (FIG. 5) superficial, unilocular, 15-40 μm diam, upper wall composed of one layer of brown-walled cells radiating from the center, lower wall lacking, base consisting of a brown pellicle. Ostiole consisting of a cell wall forming a tube up to 8 μm tall (FIG. 5), approximately in the middle of a conidioma. Conidiophores absent. Conidiogenous cells (FIG. 5) phialidic, determinate, discrete, hyaline, smooth, making up the wall. Conidia bacillar, (FIG. 5) 2.4-3.2 x 0.5-0.8 μm, aseptate, hyaline, thin-walled, smooth.

Microthyrium arizoniensis A. W. Ramaley, *sp. nov.*

Mycelium superficiale brunneum, septatum, ramosum, hyphopodiis, mycelium internum in cellulis epidermalibus, parenchymatibus, fibris. Ascomata superficialis

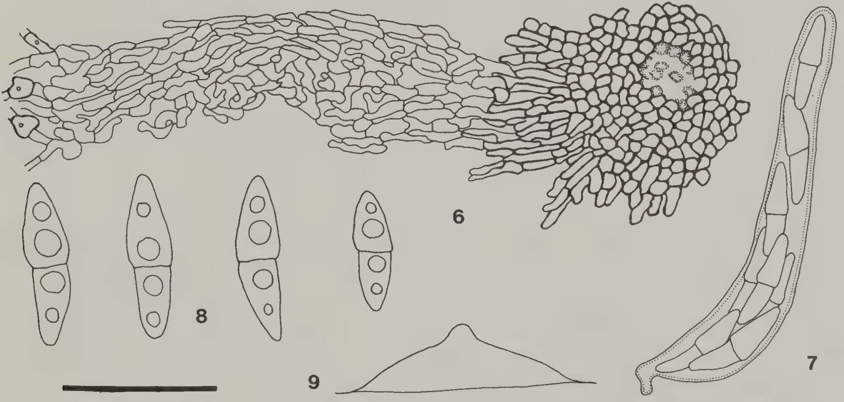


FIGS. 1-5. *Microthyrium nolinae* (AWR 9524). FIG. 1. Surface view of part of an ascoma. FIG. 2. Median longitudinal section of ca. half an ascoma. FIG. 3. Ascospores stained with cotton blue. FIG. 4. Detail of asci and pseudoparaphyses. FIG. 5. Median longitudinal section and surface view of a conidioma accompanying *M. nolinae* ascomata. Standard line: 1, 2=36.5 μm; 3=12.5 μm; 4=20 μm; 5=18 μm.

prope basim foliorum emortuorum, scutata, 125-425 μm diam, papillata, supra peridium ca 8-10 μm crassum, ex 3 vel 4 stratis cellularum parvarum, brunnearum, aliquantum crassitunicatarum, stratum basale tenue, cellulae subbrunneae, tenuitunicatae. Hamathecium filis ramosis, cellulosis, inter ascos, periphyses praesentes. Asci numerosi, bitunicati, obclavati vel prope cylindracei, leviter inequilaterales, 45-64 x 6.4-8.0 μm, ascosporis 8 irregulariter dispositis. Ascosporae 12-14.4 x 3.2-4.0 μm, 1-septatae, hyalinae, tenuitunicatae, plerumque guttulae 2, magnae utraque cellula. Appendiculae non visae.

HOLOTYPUS: in foliis emortuis Nolina microcarpa Wats. ”U. S. A. ARIZONA: Gila County, State Hwy 87, 0.56 km S of mile 224, E side of road, 8 April 1996” *a Annette Ramaley n. 9601 lectus, in BPI depositus. Paratypus in foliis emortuis Nolina microcarpa* Wats. ”U. S. A. ARIZONA: Yavapai County, E side of Interstate 17, mile 300.7, 4 April 1996” *a Annette Ramaley n. 9603 lectus, in UC depositus.*

Superficial mycelium brown, septate, branched, appressed to the substrate, with hyphopodia, internal mycelium in epidermal, parenchyma, and fiber cells, connected to bottom layer of wall and superficial mycelium by thin hyphae through epidermal cell wall and cuticle. Ascomata (FIG. 9) superficial near the base of dead leaves, scutate, outline approximately circular, separate or sometimes confluent, when confluent each with its own ostiole, 125-425 μm diam, dark, centrally more or less papillate, ostiole 12-25 μm diam; upper peridium ca 8-10 μm thick, composed of 3 or 4 layers of small, brown, somewhat thick-walled cells, basal layer thin, composed of small, pale brown,



FIGS. 6-9. *Microthyrium arizoniensis* (AWR 9601). FIG. 6. Surface view of part of an ascoma. FIG. 7. Ascus. FIG. 8. Ascospores. FIG. 9. Side view of an ascoma. Standard line: 6=36.5 μm; 7=20 μm; 8=20 μm; 9=143 μm.

thin-walled cells; peridium surface (FIG. 6) of more or less radiating rows of cells, ca isodiametric and thicker walled in the papilla, becoming more elongate, thinner walled, and less regular in shape away from the papilla. Hamatheciium of branched, cellular filaments between the asci, persistent, periphyses present. Asci (FIG. 7) numerous, maturing successively, obliquely directed toward the pore, bitunicate, obclavate to nearly cylindric, slightly inequilateral, 45-64 x 6.4-8.0 μm, with 8 ascospores, most maturing at the base but often with 1-3 ascospores in the neck. Ascospores (FIG. 8) 12-14.4 x 3.2-4.0 μm, 1-septate, hyaline, thin-walled, each cell usually with 2 large guttules. Ciliary appendages not seen.

Microthyrium guadalupensis A. W. Ramaley, *sp. nov.*

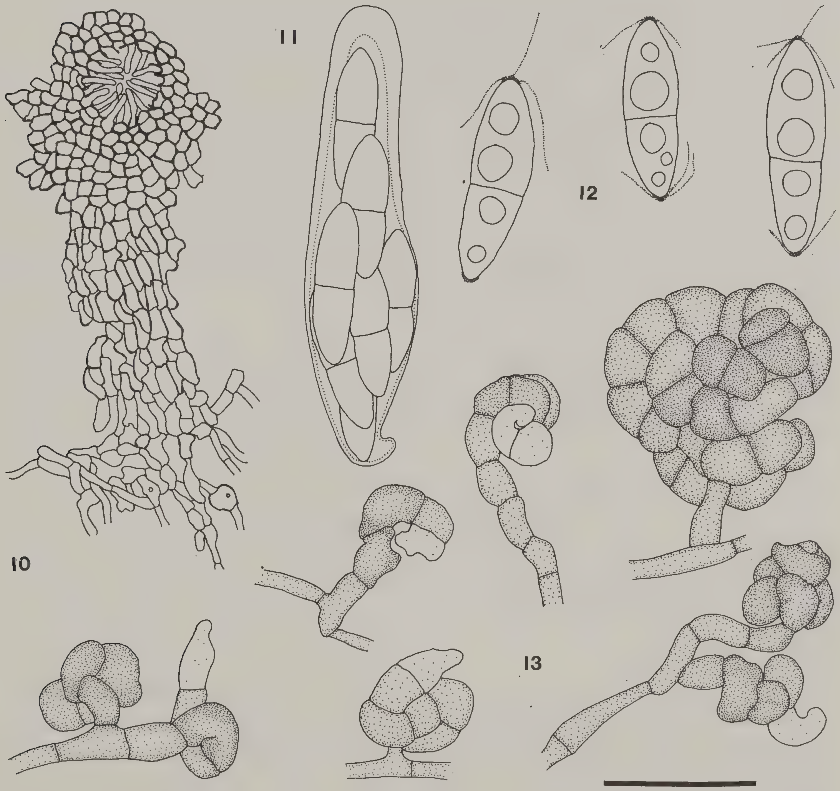
Mycelium superficiale brunneum, septatum, ramosum, hyphopodiis, mycelium internum in cellulis epidermalibus, parenchymatibus, fibris. Ascomata superficiale prope basin foliiorum emortuorum, scutata, 200-375 μm diam, supra peridium ca 15-20 μm crassum, ex 4 vel 5 stratis cellularum parvarum, brunnearum, aliquantum crassitunicatarum, interdum complanatarum constans, stratum basale tenue, cellulae parvae, subbrunneae, tenuitunicatae. Hamathecium filis ramosis, cellulosis, inter ascos; periphyses et periphysoides praesentes. Asci numerosi, bitunicati, obclavati, leviter inequilaterales, 48-68 x 12-15.2 μm, ascosporis 8 vel interdum 4. Ascosporae 15.2-19.2(-21.6) x 4.8-5.6(-6.4) μm, 1-septatae medianae vel leviter submedianae, hyalinae, tenuitunicatae, plerumque guttulae 2 magnae utraque cellula. Ascosporae nodo ad basim et apicem, nodi appendicibus subtilibus 1-4, usque ad 10μm longis sed plerumque brevioribus, saepae effractis vel imperfecti formatis.

HOLOTYPUS in foliis emortuis Nolina micrantha I. M. Johnst., "U. S. A. TEXAS: Culberson County, Guadalupe Mountains National Park, McKittrick Canyon, 17 May 1995" *a Annette Ramaley n. 9501 lectus, in BPI depositus.*

Superficial mycelium brown, septate, branched, appressed to the substrate, with hyphopodia; internal mycelium in epidermal, parenchyma, and fiber cells, connected to bottom layer of wall and superficial mycelium by thin hyphae through epidermal cell wall and cuticle. Ascomata superficial near the base of dead leaves, scutate, outline approximately circular, separate or rarely confluent, when confluent each with its own ostiole, 200-375 μm diam, dark, with a central ostiole up to 30 μm diam, usually narrower; upper peridium ca 15-20 μm thick, composed of 4 or 5 layers of small, brown, somewhat thick-walled, sometimes flattened cells, basal layer thin, cells small, pale brown, thin-walled; surface of peridium (FIG. 10) of more or less radiating rows of cells, ca isodiametric and thicker walled in 4-6 layers around the ostiole, becoming somewhat more elongate and thinner walled away from the ostiole. Hamathecium of branched, cellular filaments between the asci, persistent; periphyses and periphysoids present. Asci (FIG. 11) numerous, maturing successively, obliquely directed toward the pore, bitunicate, obclavate, slightly inequilateral, upper wall often quite thick, 48-68 x 12-15.2 μm, with 8 or occasionally 4 ascospores. Ascospores (FIG. 12) 15.2-19.2(-21.6) x 4.8-5.6(-6.4) μm, medianly or slightly submedianly 1-septate, hyaline, thin-walled, each cell usually with 2 large guttules. Ascospores with a node at both apex and base, each node bearing 1-4 very delicate ciliary appendages, up to 10 μm long but generally shorter, often broken off or imperfectly formed.

Anamorph: *Zalerion guadalupensis* A. W. Ramaley *sp. nov.*

Ex cultura cognitus. Mycelium brunneum; hyphae plerumque 2.5-4.0 μm latae, ramosae, septatae. Coloniae brunneae, velutinae, hyphis brevibus erectis et mycelio copioso in agaro. Conidia plerumque ad paginam et subpaginam agari, aliquot in



FIGS. 10-13. *Microthyrium guadalupensis* (AWR 9501). FIG. 10. Surface view of part of an ascoma. FIG. 11. Ascus. FIG. 12. Ascospores. FIG. 13. Conidia of *Zalerion guadalupensis*, the anamorph of *M. guadalupensis* produced in culture. Standard line: 10=36.5 μm; 11, 13=20 μm; 12=12.5 μm.

hyphis erectis, formata. Cellulae conidiogenae laterales vel terminales in ramis brevis, plerumque similiter hyphis reliquo mycelii, interdum conidia lateralia approximata in ramis ex cellulis hyphis usitatis brevioribus, latioribus, brunnioribus constantibus formata. Conidia solitaria, primo plus minusve helicoidea, filum multiseptatum, ramosum, irregulariter diversum circinnatum et plerumque nodos cellularum formans, nodi interdum coalescentes et massa usque ad admodum ca 130 x 60 μm formantes, cellulae conidiorum saepe irregulariter lobatae.

HOLOTYPUS in foliis emortuis Nolina micrantha I. M. Johnst., "U. S. A. TEXAS: Culberson County, Guadalupe Mountains National Park, McKittrick Canyon, 17 May 1995" *a Annette Ramaley n. 9501 lectus, ex culto agaro sicco in BPI depositus.*

Known only from culture. Mycelium brown; hyphae mostly 2.5-4 μm wide, branched, septate. Colonies brown, velvety, with short upright hyphae and abundant mycelium in the agar. Most conidia formed at the agar surface and below, a few on the upright hyphae. Conidiogenous cells lateral or terminal on short branches, mostly on hyphae similar to the rest of the mycelium, sometimes closely spaced lateral conidia formed on branches with shorter, broader, darker cells than the usual hyphal cells. Conidia solitary, more or less helicoid at first (FIG. 13), filament multiseptate, branched, irregularly coiled in several directions and generally forming knots of cells (FIG. 13) which may coalesce with adjacent conidial knots to form large masses up to at least ca 130 x 60 μm, conidial cells often irregularly lobed.

Conidium formation and colony characters are as in *Zalerion* Moore & Myers (1962), emend Anastasiou (1963), a genus common in submerged wood in saline environments. A nonaquatic species has been described from cankers of *Larix decidua* and stem wounds of *Picea abies* in Great Britain (Buczacki 1972).

Coelomycetes resembling that found with *Microthyrium nolinae* also accompanied *M. arizoniensis* and *M. guadalupensis* but were not described because some characters could not be satisfactorily ascertained. The coelomycete is most noticeable near younger ascomata. Conidia are the size of many ascomycete spermatia, and their proximity to young ascomata suggests such a role but is not decisive.

The 3 new *Microthyrium* species are distinct from those from the United States described in modern terms (Barr 1990) as well as 3 other species listed in Farr *et al* (1989). The description for *M. michelianum* Togn. lacks detail, but the host (*Castanea)* and ascospore size and shape make it an unlikely synonym for any of the new species. *Microthyrium rubicola* H. C. Greene is astomate and is therefore excluded from the genus. *Microthyrium mitchellae* Cooke is problematic. The species is listed in *Index of Plant Diseases in the United States* (Anonymous 1960), *Index of Plant Diseases in Florida* (Alfieri *et al* 1984), and *Fungi on Plants and Plant Products in the United States* (Farr *et al* 1989), but an original description could not be located. Dr. Tim Schubert, Florida Department of Agriculture and Consumer Services, provided an explanation--the species is most likely an erroneous citation. The entry for *M. mitchellae* on page 48 of *The North American Pyrenomycetes* (Ellis and Everhart 1892) was probably mistaken for a *Microthyrium* species (page 44) instead of a *Meliola* species (page 45), for the change in genus is easily overlooked. The error apparently originated in compiling the 1960 USDA index, from which species were accepted for the first plant disease index of Florida, and has been perpetuated in the literature since that time.

At least 11 species in addition to *Microthyrium macrosporum* have been found to have ascospore appendages. In most species the appendages are apical or apical and septal. No northern hemisphere species described in modern terms has apical and basal appendages as found in *M. guadalupensis*. A globose or penicillate appendage, whose structure is not well detailed, was reported on both ends of **young** ascospores of *M. pulchellum* Speg. (cited in Saccardo 1891). This Brazilian fungus is from living *Blechnum* leaves and has smaller ascomata and asci than *M. guadalupensis*, so the species are not likely synonyms. As in *M. nolinae*, ascospores of *M. ciliatum* Gremmen & de Kam, *M. gramineum* Bomm., Rouss., & Sacc., and *M. pinophyllum* have lateral appendages on the upper hemispore. However, these 3 species have 2 tufts of 3 appendages connected by a short thread rather than up to 20 delicate, radially disposed appendages from a common node. For *M. nolinae* the outermost wall layer is stainable and continuous in young ascospores. The stainable material appears as helically arranged strips on older ascospores. At maturity and release from the ascus, the stainable strips spring outward forming the divergent appendages. These observations suggest that *M. nolinae* ascospore appendages originate in a manner similar to conidial appendages designated Type D (Nag Raj 1993), or perhaps to the equatorially attached appendages of *Halosphaeria mediosetigera* Cribb & Cribb (Moss and Gareth-Jones 1977).

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My thanks to Margaret Barr for reviewing the manuscript, and to Guadalupe Mountains National Park for permission to collect within the park. Thanks are also due Margaret Barr for help with the literature, and Tim Schubert whose detective work provided an explanation for the lack of an original description for *Microthyrium mitchellae*!

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NEW SMUT FUNGI FROM SOUTH AFRICA

KÁLMÁN VÁNKY

Herbarium Ustilaginales Vánky (HUV)
Gabriel Biel Str. 5, D-72076 Tübingen, Germany

ABSTRACT

NEW GENUS proposed: *Ustanciosporium* K. Vánky, type *U. rhynchosporae* K. Vánky, sp. nov. (on *Rhynchospora rugosa*, Cyperaceae).

NEW SPECIES proposed: *Aurantiosporium marisci* K. & C. Vánky (type on *Mariscus thunbergii*, Cyperaceae), *Tolyposporium epischoeni* K. Vánky (type on *Epischoenus gracilis*, Cyperaceae), *Sporisorium pole-evansii* K. Vánky (type on *Digitaria eriantha* and *D. monodactyla*, Gramineae), and *Ustilago drakensbergiana* K. Vánky (type on *Digitaria tricholaenoides*, Gramineae).

Key words: smut fungi, new taxa, South Africa.

The study of smut fungi, recently collected in South Africa, resulted in the recognition of several new taxa.

Aurantiosporium marisci K. & C. Vánky, **sp. nov.**

Typus in matrice *Mariscus thunbergii* (Vahl) Schrad. (*M. riparius* Schrad., Cyperaceae; det. C. Archer, PRE), South Africa, Western Cape Prov., Tsitsikamma National Park, Nature's Valley, 33°58'37" S, 23°33'45" E, alt. cca. 2 m, 19.XII.1996, leg. C. & K. Vánky. Holotypus in Herbario Ustil. Vánky (HUV 18031!), isotypi in PREM, BPI et in Vánky, Ust. exs. No. 1001.

Sori in spiculis, primo circuli parvi, aperti ad basin omnium glumarum, axem floralem circumdantes, diametro 0,5–1,5 mm, flavidobrunnei, strato tenui epidermidis hospitis cooperti, quo irregulariter rupto massam flavidobrunneam usque rubiginosam, granulosam-pulveream glomerulorum sporarum ostendentes. Infectio systemica; spiculae et flores omnes eiusdem inflorescentiae affectae.

Fungus proliferationem validam spiculae producens, illam 10–20 mm longam et 20–30 vel plus flores habentem, loco illae integrae cca. 7 mm longae et 3 flores habentis, producens. Maturatio sororum a floribus basalibus distaliter progrediens. Glumae sororum maturorum facile deciduae et axis spicularum denudatus cum residuis sororum manens. *Glomeruli sporarum* forma et magnitudine varii, irregulares, elongati, raro subglobosi, 25–80 x 30–120 µm, satis persistentes, e sporis (1–)2–25(vel plus), citrinoflavis usque atro-aurantiacis compositi. *Sporae* forma et magnitudine valde variae, subpolyedriciter irregulares vel elongatae, raro subglobosae vel ellipsoidales, saepe fusiformes, in rostra 1 vel 2 subacuta prolongatae, 13–25(–28) x 16–60(–65) µm (rostrum inclusum); pariete levi, multistratoso, crassitudine valde vario, endosporium regulare, cca. 0,5 µm crassum, exosporium 1,5–10 µm vel ad rostrum usque 25 µm crassum.

Sori (Fig. 1) in the spikelets, at first as small, swollen, open rings at the base of each glume, surrounding the floral axis, 0.5–1.5 mm in diameter, yellowish-brown, covered by a delicate layer of host epidermis which ruptures irregularly, disclosing the yellowish- to rusty-brown, granular-powdery mass of spore balls. Infection systemic; all spikelets and flowers of an inflorescence are affected. The fungus produces a strong proliferation of the spikelets which become 10–20 mm long with up to 20–30 or more flowers instead of the c. 7 mm length and three flowers of the healthy spikelets. The sori ripen from the basal flowers, progressing distally. The glumes of the ripe sori detach easily and the axis of the spikelets becomes naked, with soral remnants. *Spore balls* (Figs. 3, 4) variable in shape and size, irregular, elongated, rarely subglobose, 25–80 x 30–120 µm, rather persistent, composed of (1–)2–25(or more?), lemon-yellow to dark orange-yellow spores. *Spores* (Figs. 3, 4) very variable in shape and size, subpolyhedrally irregular or elongated, rarely subglobose or ellipsoidal, often fusiform, prolonged into one or two subacute rostra, 13–25(–28) x 16–60(–65) µm (including rostrum); wall smooth, multilayered, very variable in thickness, endospore regular, c. 0.5 µm thick, exospore 1.5–10 µm wide or up to 25 µm at the rostrum.

The monospecific genus ***Aurantiosporium*** M. Piepenbring, K. Vánky & Oberwinkler (1996:62) was erected for *Ustilago subnitens* Schröt. & Henn., type on *Scleria pratensis* Lindley ex Nees (= *S. melaleuca* Reichb. ex Schldl. & Cham., Cyperaceae) from Brazil. New data and the discovery of a second, closely related species, allow a completed description of this genus:

Sori in spikelets of Cyperaceae producing hypertrophy. Spore mass orange-yellow to rusty-brown, granular-powdery, composed of loose or permanent spore balls. Spores irregular, pigmented (yellow to yellowish-red). Spore wall of irregular thickness, multilayered. Spore germination results in phragmobasidia with sessile basidiospores. Host–parasite interaction by intercellular hyphae. Septa with simple pores lacking tripartite caps.

In the new classification, *Aurantiosporium* belongs to the Ustilentylomataceae R. Bauer & Oberw., of the Order Microbotryales R. Bauer & Oberw. (Bauer et al., 1997:1307-1311), within the Class Uredinomycetes (Begerow et al., 1997).

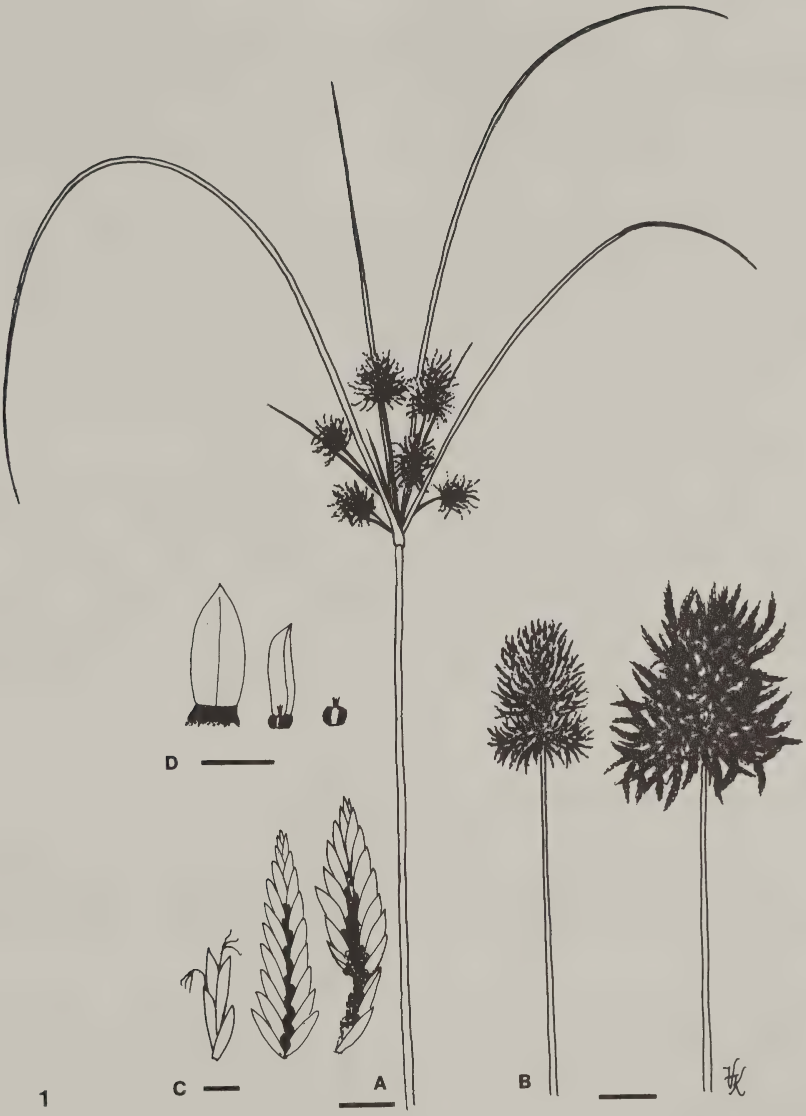


Fig. 1. *Aurantiosporium marisci* K. & C. Vánky on *Mariscus thunbergii* (Vahl) Schrad. **A**. Infected inflorescence. **B**. Healthy and an infected part-inflorescence; the infected one with elongated, many-flowered spikelets. **C**. Healthy and two infected spikelets, showing the same phenomenon, the strong proliferation of the infected spikelets. **D**. Glume with remnants of a ripe sorus at its base, and two unripe sori, one with removed glume (type).
Bar of A = 5 cm, B = 1 cm, C and D = 2 mm.

Tolyposporium epischoeni K. Vánky, **sp. nov.**

Typus in matrice *Epischoenus gracilis* Levyns (Cyperaceae; det. C. Archer, PRE) South Africa, Eastern Cape Prov., Bloukrans Pass, 20 km W. oppid. Stormsrivier, via No. 102, 33°57'44" S, 23°42'00" E, alt. cca. 280 m, 19.XII.1996, leg. C. & K. Vánky. Holotypus in Herbario Ustil. Vánky (HUV 18035), isotypi in PREM et in BPI.

Sori in spiculis parum tumefactis, glumis perfecte occulti, massa nigra, granuloso-pulverea glomerulorum sporarum circa organa floralia interna productorum impleti. *Glomeruli sporarum* subglobosi, ovoidei, elongati vel irregulares, 35–100 x 40–180 µm, nigri, opaci, permanentes, e multitudine sporarum compositi. *Sporae* atrobrunneae, visu mediano subcuneiformes, elongatae vel irregulares, 8–28 µm longae, visu superficiali irregulariter subpolygonales, 5–10 x 6–15 µm; pariete libero parum convexo, 1,5–2,5(–3) µm crasso, irregulariter verrucoso atque residuis hypharum sporogenarum facile separabilium; pariete lateris contactus tenui (cca. 0,5 µm), levi.

Sori (Fig. 2) in the slightly swollen spikelets completely hidden by the glumes, filled with a black, granular-powdery mass of spore balls produced around the internal floral organs. *Spore balls* (Figs. 5, 6) subglobose, ovoid, elongated or irregular, 35–100 x 40–180 µm, black, opaque, permanent, composed of a great number of spores. *Spores* (Fig. 5) blackish-brown, in median view subcuneiform, elongate or irregular, 8–28 µm long, in surface view irregularly subpolygonal, 5–10 x 6–15 µm; free wall slightly convex, 1.5–2.5(–3) µm thick, irregularly verrucose and also with remnants of sporogenous hyphae which become detached easily; wall of contact sides thin (c. 0.5 µm), smooth.

TWO NEW SMUT FUNGI ON *DIGITARIA* (GRAMINEAE)

To be able to identify some smut fungi collected on *Digitaria*, I revised the Ustilaginomycetes of this grass genus (Vánky, in preparation). Nine species could be distinguished. Two additional species from South Africa turned out to be new.

I. B. Pole-Evans (1879-1968) collected a smut fungus on *Digitaria monodactyla* in South Africa, and deposited it in PREM (No. 9416!) under the name *Ustilago inconspicua* Pole-Evans (nom. herb.). Zundel (1938:304), studying the smut fungus collection in Pretoria, discovered this species and described it as "*Sorosporium inconspicuum* (Pole-Evans) Zundel, n. n.", without giving a Latin diagnosis (invalid name, ICBN 36.1). This fungus was collected several times in South Africa, also on other *Digitaria* species. It shows characters of a *Sporisorium*. [The name of *Sporisorium inconspicuum* is already occupied for another fungus, *S. inconspicuum* (Zundel) K. Vánky, based on *Sphacelotheca inconspicua*, type on *Mesosetum loliiforme* (Hochst.) Chase, Brazil]. It is described as:

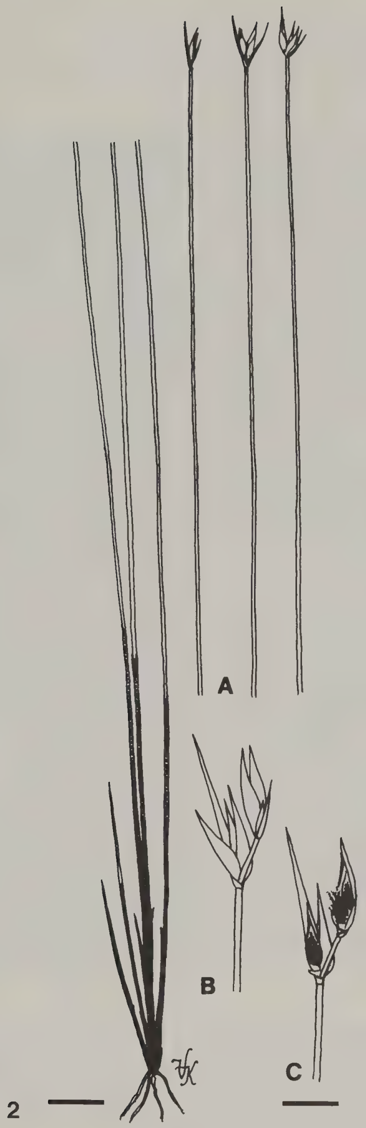
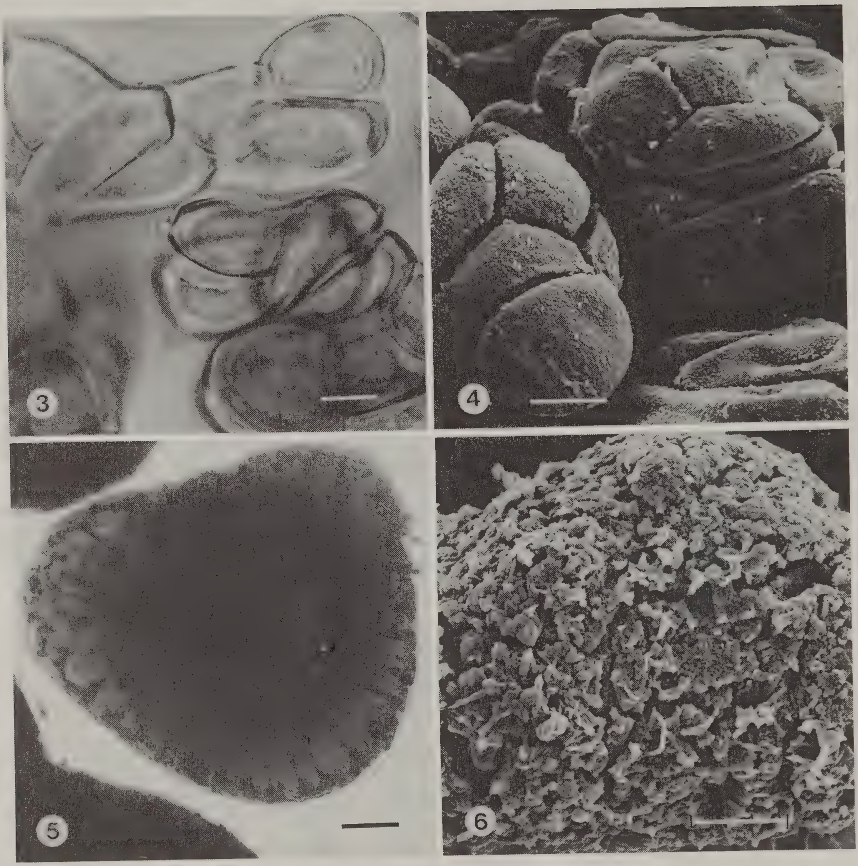


Fig. 2. Sori of *Tolyposporium epischoeni* K. Vánky in the somewhat swollen spikelets of *Epischoenus gracilis* Levyns. **A**. Two infected inflorescences and, to the left, a healthy one. An infected inflorescence, **B**. with glumes, **C**. the same with partly removed glumes, with the masses of spore balls produced around the inner floral organs (type). Bar of A = 1 cm, B and C = 5 mm.



Figs. 3, 4. Spore balls and spores of *Aurantiosporium marisci* K. & C. Vánky on *Mariscus thunbergii* (Vahl) Schrad. in LM and in SEM (type).

Figs. 5, 6. Spore balls and spores of *Tolyposporium epischoeni* K. Vánky on *Epischoenus gracilis* Levyns in LM and in SEM. The dark, compact spores within the spore balls are hardly visible (type). Bars = 10 µm.

Sporisorium pole-evansii **K. Vánky, sp. nov.**

Typus in matrice *Digitaria eriantha* Steud., South Africa, Gauteng Prov., cca. 15 km S. urbe Pretoria, pr. Irene Institute, 25°53'52" S, 28°12'42" E, alt. 1500 m, 15.I.1997, leg. C. Roux, C. & K. Vánky. Holotypus in Herbario Ustil. Vánky (HUV 18452!), isotypi in PREM, BPI et in Vánky, Ust. exs. No. 1026. Paratypus in matrice *Digitaria monodactyla* (Nees) Stapf, South Africa, KwaZulu-Natal Prov., Drakensberg Mts., Cathedral Peak National Park, Mikes Pass, 28°58'07" S, 29°19'04" E, alt. c. 2000 m, 30.XII.1996, leg. C. & K. Vánky (HUV 18453!), isoparatypi in PREM et in BPI.

Ustilago inconspicua Pole-Evans (nom. herb.). — *Sorosporium inconspicuum* (Pole-Evans) Zundel, 1938:304 (invalid name, no Latin diagnosis). — On *Digitaria monodactyla* (Nees) Stapf, South Africa, Pretoria, 26.XII.1915, I. B. Pole-Evans (PREM 9416!). [Not *Sporisorium inconspicuum* (Zundel) K. Vánky, based on *Sphacelotheca inconspicua*, type on *Mesosetum loliiforme*].

Sori in omnibus ovariis eiusdem inflorescentiae, plerumque singuli vel bini per spiculas, inconspicui, inter involucrum florale expansum sese ostendentes, ovoidei, cca. 1 x 2–3 mm, primo peridio crasso, pallide brunneo cooperti, illo ab apice disrupto massam atro-olivaceobrunneam, pulveream sporarum et columellam unam centralem simplicem, vel in apice breviter bi- vel trifurcatam ostendentes. Infectio systemica, plantae infectae et inflorescentiae earum humiliores. *Sporae* maturae singulae, globosae, ovoideae, ellipsoidales, plerumque parum irregulares, rotundato-subpolyedricae, 6,5–9 x 7–10(–11) μm, flavidobrunneae; pariete aequali, cca. 0,5 μm crasso, modice dense humiliter verrucoso-echinulato, lineam extremam sporae vix efficienti eam leniter serrulatam apparentem ostendenti. *Cellulae steriles* subglobosae, ovoideae, cylindricae vel irregulares, 5–9 x 5–13 μm, singulae, in globis parvis vel in catenis, hyalinae, leves; pariete tenui (cca. 0,3 μm). *Germinatio sporarum* cum basidiis septatis, 2,5 x 20–33 μm. Cellulae basidiales vicinae plerumque per pontem copulationis brevem confusae hypham vel etiam basidiosporas breviter vel longe ellipsoidales, 1–1,5 x 3–13(–18) μm, successive germinantes evolventes.

Sori (Fig. 7) in all ovaries of an inflorescence, usually one, rarely two per spikelet, inconspicuous, showing between the spreading floral envelopes, ovoid, c. 1 x 2–3 mm, first covered by a thick, light brown peridium which soon ruptures from its apex disclosing the dark olivaceous-brown, powdery mass of spores and a central columella, simple or shortly bi- or trifurcate on its apex. Infection systemic, infected plants and inflorescences shortened. *Spores* (Figs. 11, 12) when mature single, globose, ovoid, ellipsoidal, usually slightly irregular, rounded subpolyhedral, 6.5–9 x 7–10(–11) μm, yellowish-brown; wall uniform, c. 0.5 μm thick, moderately densely low verrucose-echinulate just affecting the spore profile which appears finely serrulate. *Sterile cells* subglobose, ovoid, cylindrical or irregular, 5–9 x 5–13 μm, single, in small groups or in chains, hyaline, thin-walled (c. 0.3 μm), smooth. *Spore germination* (Fig. 8; on water-agar, at room temp., after 2 days) results in septate basidia measuring 2.5 x 20–33 μm. The adjoining basidial cells usually fuse through a short copulation

bridge giving rise to hyphae but also to short or long ellipsoidal basidiospores of 1–1.5 x 3–13(–18) µm, which bud successively.

On Gramineae: *Digitaria argyrograpta* (Nees) Stapf, *D. eriantha* Steud. (*D. pentzii* Stent), *D. monodactyla* (Nees) Stapf, and *D.* sp., South Africa and Malawi.

Differences of up to 1 µm in spore measurements were noticed between samples from various host plant species. However, because in all other aspects these samples were identical, I am considering these differences to be variations within the same species.

Ustilago drakensbergiana K. Vánky, **sp. nov.**

Typus in matrice *Digitaria tricholaenoides* Stapf, South Africa, KwaZulu-Natal Prov., Drakensberg Mts., Cobham Nature Reserve, pr. Camp site, 29°41'58" S, 29°41'45" E, alt. 1620 m, 7.I.1997, leg. C. & K. Vánky. Holotypus in Herbario Ustil. Vánky (HUV 18456!), isotypi in PREM et in Vánky, Ust. exs. No. 1046.

Sori in omnibus axibus racemorum inflorescentiae eiusdem, longe cylindrici, irregulares, recurvati, 1–2,5 mm lati, 3–8 cm longi, saepe spiculas parvas abortivas formantes, primo membrana tenui, argentea, origine matricali cooperti, qua in partibus nonnullis irregulariter disrupta, massam pulveream, atro-olivaceobrunneam sporarum et residua telarum vascularium sicut "columellas" longas, filiformes ostendentes. Neque glomeruli sporarum, neque cellulae steriles. *Sporae* globosae, subglobosae, ovoideae vel ellipsoidales, (4–)4,5–6 x 5–6,5(–7) µm, flavidobrunneae, in uno latere parum pallidiores; pariete tenui (cca. 0,4 µm), inconspicue punctato-verrucoso. *Germinatio sporarum* cum basidiis septatis basidiosporas ovoideas usque ellipsoidales 1,5 x 3–7 µm, et hyphas septatas, ramificatas, cca. 1,5 µm crassas productis. Ad hyphas forte sporae basidiosporis similes pullulantes productae.

Sori (Fig. 9) in all raceme axes of an inflorescence, long cylindrical, irregular, recurved, 1–2.5 mm wide, 3–8 cm long, often bearing small, aborted spikelets, first covered by a thin, silvery membrane of host origin which ruptures irregularly at several places disclosing the dark olivaceous-brown, powdery mass of spores and remnants of vascular tissues as long, filiform "columellae". No spore balls, no sterile cells. *Spores* (Figs. 13, 14) globose, subglobose, ovoid or ellipsoidal, (4–)4.5–6 x 5–6.5(–7) µm, yellowish-brown, slightly paler on one side; wall thin (c. 0.4 µm), indistinctly punctate-verrucose. *Spore germination* (Fig. 10; on water-agar, at room temp., after 3 days) results in septate basidia on which ovoid to ellipsoidal basidiospores, measuring 1.5 x 3–7 µm, and also ramifying, septate hyphae of c. 1.5 µm diameter are produced. Basidiospore-like spores which bud may develop on the hyphae.

On Gramineae: *Digitaria tricholaenoides* Stapf, South Africa. Known only from the type collection on this endemic host plant.

U. drakensbergiana is also a *Ustilago* species which shows a more or less expressed character of the genus *Sporisorium* (presence of "columellae").

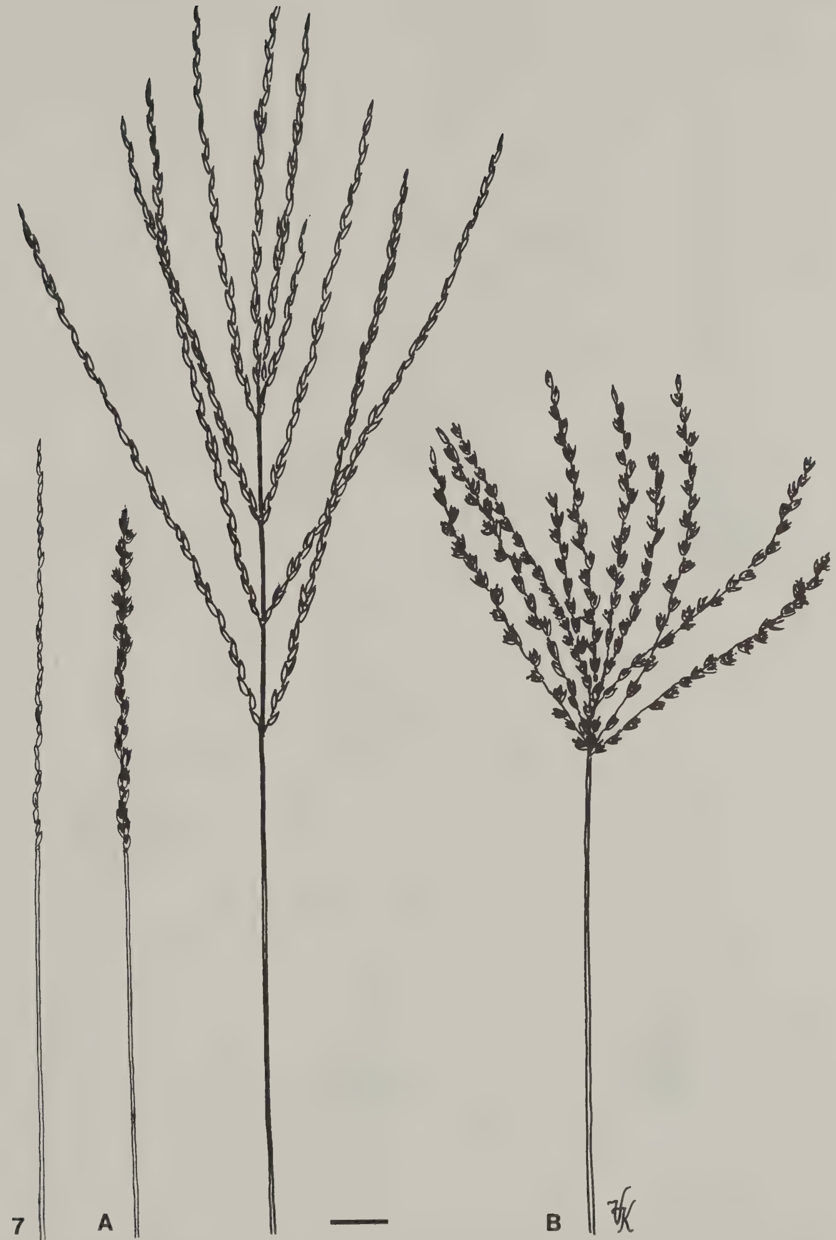


Fig. 7. Sori of *Sporisorium pole-evansii* K. Vánky in the ovaries of **A**. *Digitaria monodactyla* (Nees) Stapf (paratype), and **B**. *Digitaria eriantha* Steud. (type). Each with a healthy inflorescence to the left. Bar = 1 cm.

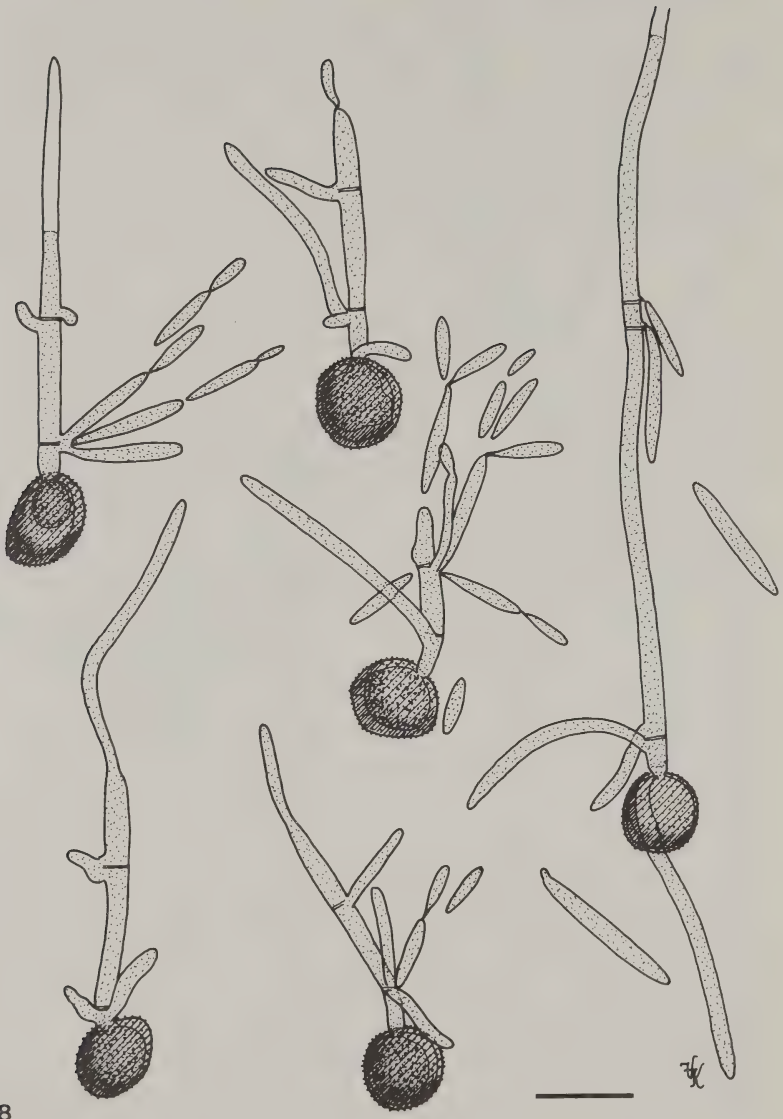


Fig. 8. Germinating spores of *Sporisorium pole-evansii* K. Vánky (on water-agar, at room temp., after 2 days) with septate basidia on which hyphae but also short or long ellipsoidal basidiospores are borne (paratype). Bar = 10 µm.

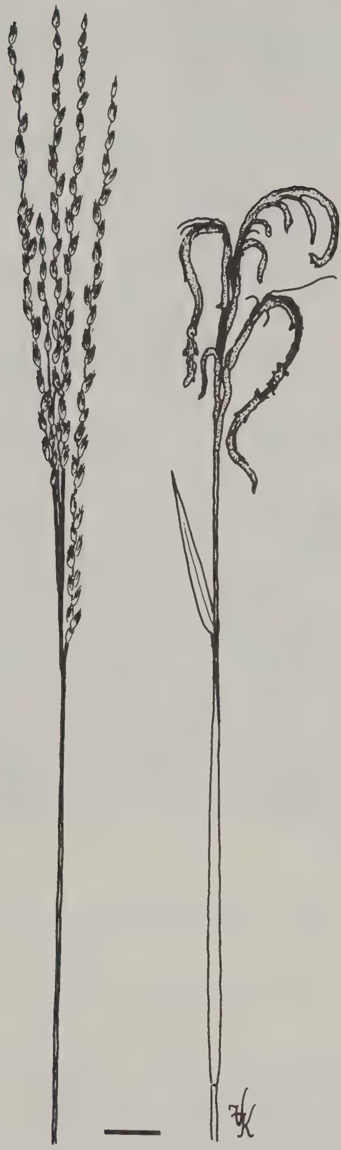


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Fig. 9. Sori of *Ustilago drakensbergiana* K. Vánky in the raceme axes of *Digitaria tricholaenoides* Stapf (type). To the left a healthy inflorescence. Bar = 1 cm.

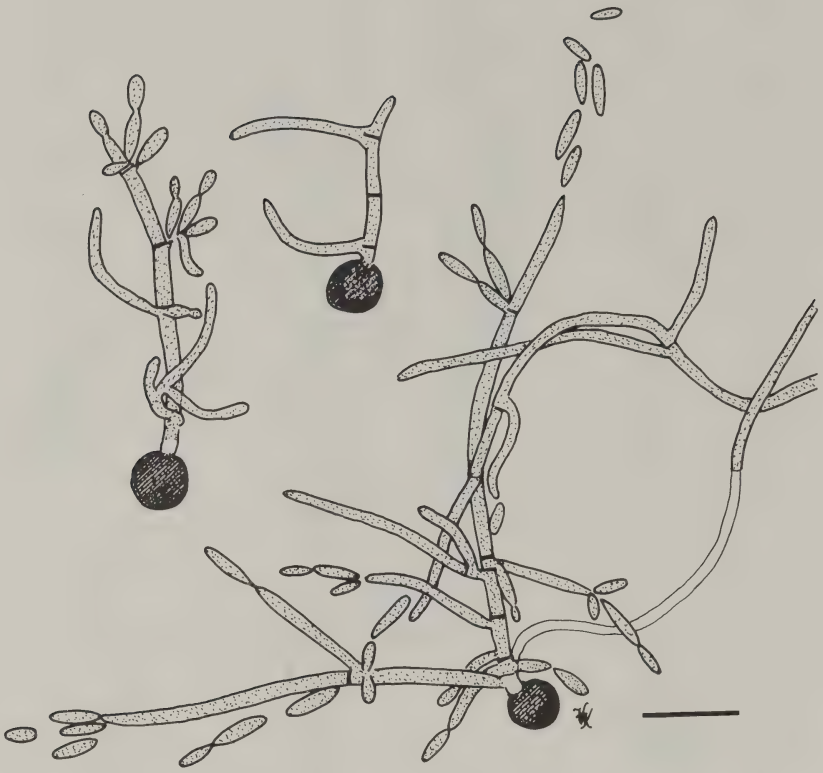

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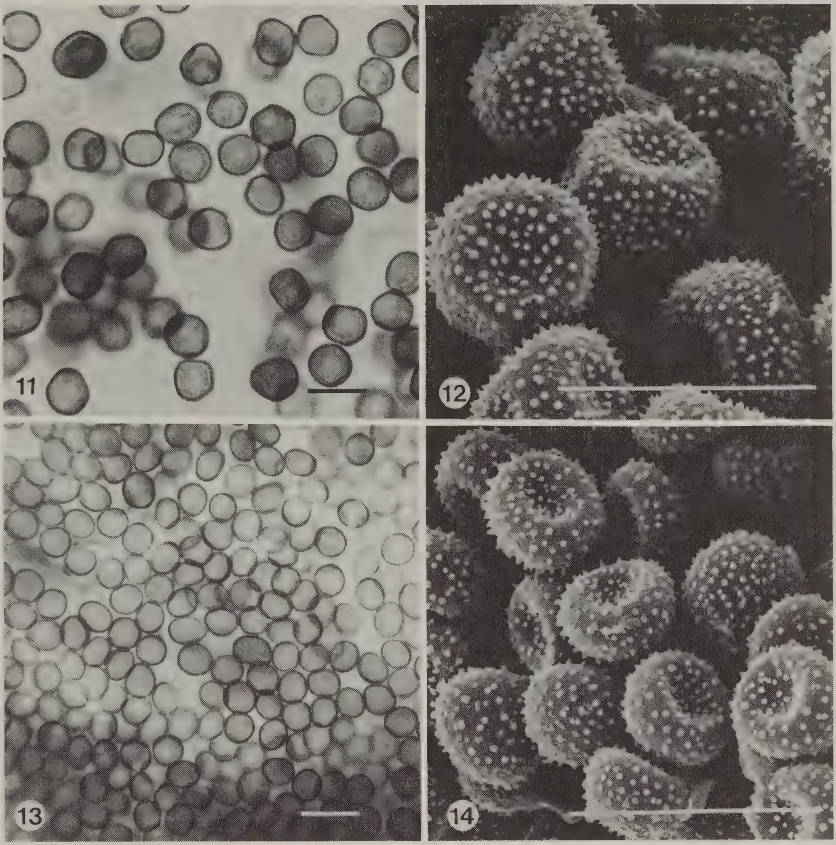
Fig. 10. Germinating spores of *Ustilago drakensbergiana* K. Vánky (on water-agar, at room temp., after 3 days) with septate basidia on which ovoid to ellipsoidal basidiospores and also ramifying, septate hyphae are produced (type). Bar = 10 µm.



Figs. 11, 12 Spores of *Sporisorium pole-evansii* K. Vánky on *Digitaria eriantha* Steud. in LM and in SEM (type).

Figs. 13, 14. Spores of *Ustilago drakensbergiana* K. Vánky on *Digitaria tricholaenoides* Stapf in LM and in SEM (type). Bars = 10 µm.

A peculiar smut fungus was collected in South Africa producing its sori in the flowers of *Rhynchospora rugosa* (Vahl) S. Gale (Cyperaceae). Its spores resemble those of the *Ustilago montagnei* Tul. & C. Tul. group, namely they are finely foveolate and possess a more or less evident, hyaline, rounded or wing-like appendage. However, whereas the *Ustilago montagnei* group has solitary spores, the recently collected fungus has spore balls. For it a new genus is proposed:

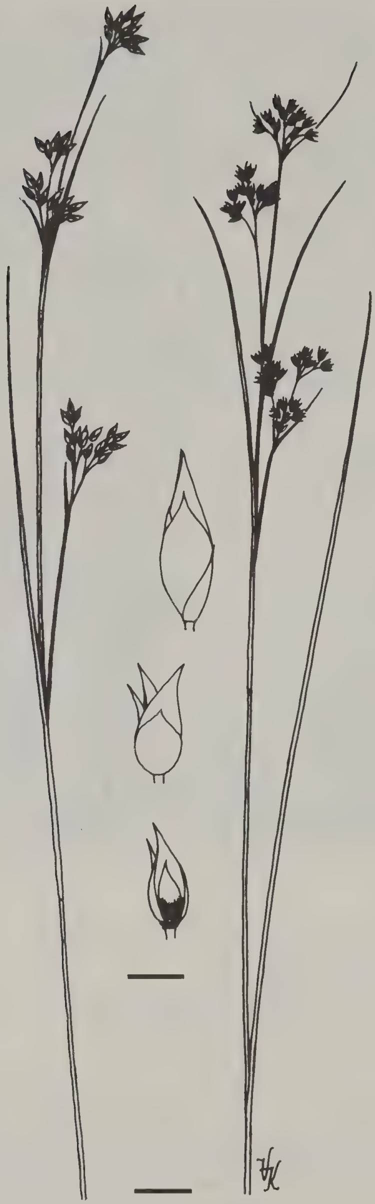


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Fig. 15. Sori of *Ustanciosporium rhynchosporae* K. Vánky in the flowers of *Rhynchospora rugosa* (Vahl) S. Gale (type). To the left a healthy inflorescence. Enlarged a healthy (top), and two diseased spikelets, one with partly removed glumes. Bars = 1 cm and 1 mm, respectively.

Ustanciosporium K. Vánky, **gen. nov.**

Sori in floribus Cyperacearum, eos massa nigra, agglutinata usque granulosopulverea glomerulorum sporarum, ad superficiem organorum floralium internorum producta implentes. *Sporae* pigmentiferae (rufobrunneae), irregulares, saepe cum appendice hyalino; pariete inaequali, ornato (foveolato).

Sori in the flowers of Cyperaceae filled with a black, agglutinated to granular-powdery mass of spore balls produced on the surface of inner floral organs. *Spores* pigmented (reddish-brown), irregular, often with a hyaline appendage; spore wall uneven, ornamented (foveolate). Typus generis:

Ustanciosporium rhynchosporae K. Vánky, **sp. nov.**

Typus in matrice *Rhynchospora rugosa* (Vahl) S. Gale, South Africa, KwaZulu-Natal Prov., Drakensberg Mts., Loteni Nature Reserve, 1 km W. Camp site, 29°26'16" S, 29°31'02" E, alt. cca. 1620 m, 6.I.1997, leg. C. & K. Vánky. Holotypus in Herbario Ustil. Vánky (HUV 18400), isotypi in PREM, BPI et in Vánky, Ust. exs. No. 1042.

Sori in floribus, a glumis externis perfecte coopertis, flores massa nigra, granuloso-pulverea glomerulorum sporarum implentes. Pars basalis sororum e telis matricalibus hyphis permeatis organorum floralium internorum plus-minus destructorum formata. Ad superficiem telarum matricalium hyphae sporogeneae in catervis parvis concentratae, ex iis glomeruli sporarum formati. Sorus ipse corpus globosum, nigrum, agglutinatum, 1–2 mm diametro, in superficie eius glomerulis maturis sporarum. Infectio systemica. Plantae infectae parum debiles, cum floribus aliquid minoribus compresse distributis. *Glomeruli sporarum* subglobosi, ovoidaei, elongati, plerumque plus-minus irregulares, 15–40 x 25–55(–70) µm, rufobrunnei, e 2–25 (vel pluribus?, plerumque 3–9) sporis compositi, compressione in sporis singulis dissoluti. *Sporae* nimis irregulares, rotundatae, subpolyedriter irregulares, elongatae, curvatae, saepe cum 1(–3) punctis subacutis vel acutis et latere uno plano vel impresso, ubi forte appendix brevis, hyalina praesens, 10–16(–18) x 14–22(–25) µm, rufobrunneae; pariete inaequali, 1–1,5 µm, ad puncta usque 3 µm crasso, leniter, satis sparse, regulariter foveolato. Germinatio sporarum ignota.

Sori (Fig. 15) in the flowers completely hidden by the outermost glumes, filled with a black, granular-powdery mass of spore balls. The basal part of the sori is formed of host tissues of the more or less destroyed inner floral organs, permeated by hyphae. On the surface of the host tissues, the sporogenous hyphae concentrate into small groups of which spore balls develop (Fig. 16). The proper sorus is a globoid, black, agglutinated body of 1–2 mm diameter, with mature, granular-powdery spore balls on its surface. Infection systemic. Infected plants slightly stunted having somewhat smaller and congested flowers. *Spore balls* (Figs. 17, 19) subglobose, ovoid, elongate, usually more or less irregular, 15–40 x 25–55(–70) µm, reddish-brown, composed of 2–25 (or more?, mostly of 3–9) spores, by pressure separating into single spores. *Spores* (Figs. 17, 18, 19) extremely irregular, rounded, subpolyhedrally irregular, elongated, curved, often with 1(–3) subacute or acute points and a flattened or impressed side, where a short, hyaline appendage can be present (arrows), 10–16(–18) x 14–22(–25) µm, reddish-brown; wall uneven, 1–1.5 µm, at the points up to 3 µm thick, finely, rather sparsely, regularly foveolate. Spore germination unknown.

On Cyperaceae: *Rhynchospora rugosa* (Vahl) S. Gale (*R. brownii* Roemer & Schultes; *R. glauca* Vahl). Known from the type collection only.

Etymology: *Ust-an-ci-o-sporium*, combined from *Ust*ilago, *An*thracoidea, *Ci*ntactia, genera with which it shows (at least apparent) similarities, and *-sporium*, the ending of many spore-ball-forming smut fungus genera.

The systematic position of *Ustilago montagnei* Tul. & C. Tul. and related species is still not satisfactorily solved. Some mycologists attributed them to *Cintractia*, others to *Ustilago*. Even the description of a new genus was considered for them (Vánky, unpublished). Ultrastructural studies (Bauer et al., 1997) did not solve this problem either. Species of *Cintractia* and *Ustilago*, including the *U. montagnei* group, all have the characters of the still heterogeneous family of Ustilaginaceae Tul. & C. Tul. emend. R. Bauer & Oberw. (containing 20 + 3? genera), namely host–parasite interaction by intracellular hyphae, and poreless mature septa. Maybe molecular biological studies, combined with classical mycological ones, will bring clarity in the classification of this group of smut fungi.

Tolyposporium Woronin ex Schröter, on Cyperaceae and Juncaceae, also belongs to the emended Ustilaginaceae. It has spore balls produced (mostly) on the surface of the host tissues, usually in the flowers. However, the morphology of the spore balls and spores, including their colour, differ markedly from those of *Ustanciosporium rhynchosporae*, and exclude a congeneric placement.

Because of the similarity of the spores of *Ustanciosporium rhynchosporae* with those of *Ustilago montagnei* (comp. Figs. 19 and 21), except for the presence of spore balls in the first species, these two species are certainly closely related; the presence or absence of spore balls does not always reflect a natural relationship. This character, however, is still a very important character in the classification of the smut fungi at the generic level (Vánky, 1998:526).

To exclude the possibility of the presence of loose spore balls, which may disintegrate early during ontogenesis, I studied the spore formation of *U. montagnei*. Spore ball building in this species could not be observed (Fig. 20).

Figs. 16–19. *Ustanciosporium rhynchosporae* K. Vánky (type).

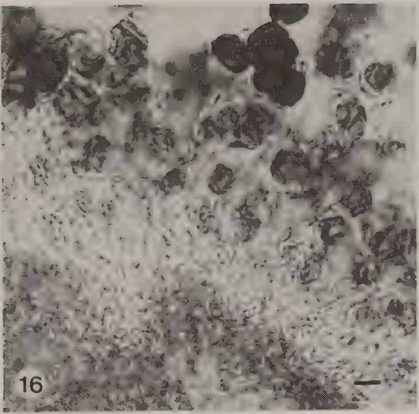
Fig. 16. Section of a young sorus, showing the development of the spore balls.

Figs. 17, 18, 19. Spore balls and spores in LM and in SEM. The arrows show the hyaline appendage of the spores in the LM picture.

Figs. 20–21. "*Ustilago*" *montagnei* Tul. & C. Tul. on *Rhynchospora alba* (Vánky, Ust. exs. No. 711, as *U. montagnei* var. *minor*).

Fig. 20. Spore formation at the basis of a young sorus. No trace of spore balls.

Fig. 21. Spores in SEM. Bars = 10 µm.



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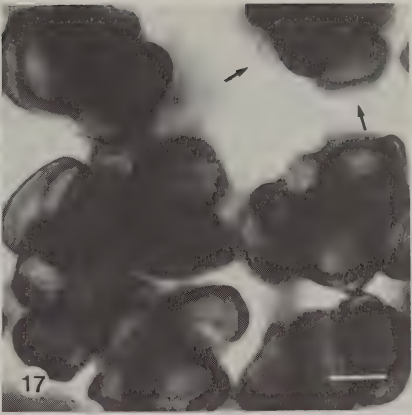

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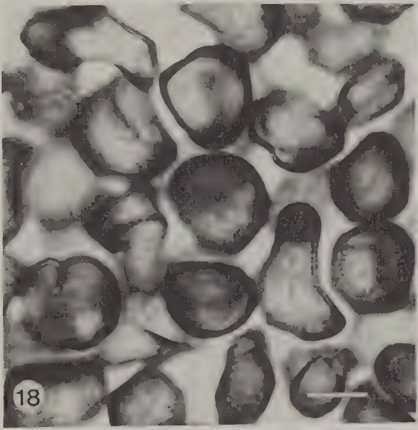

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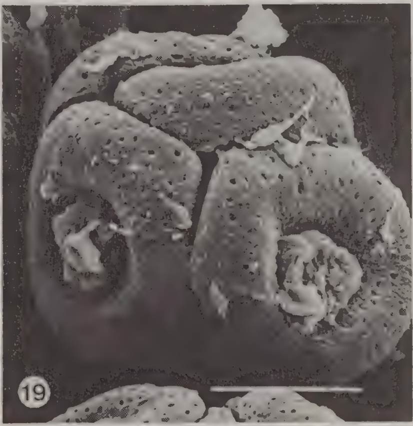

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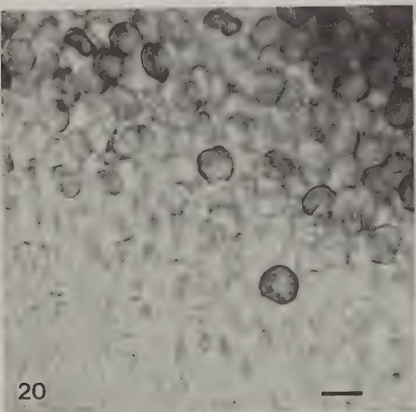

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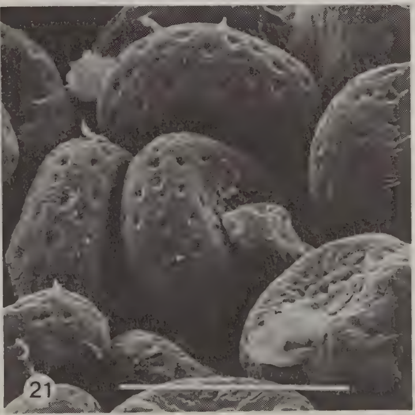

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THE NEW CLASSIFICATORY SYSTEM FOR SMUT FUNGI, AND TWO NEW GENERA

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ABSTRACT

After a short introduction and the presentation of the new classificatory system for the smut fungi and with them related taxa, **new genera** proposed are: ***Tothiella*** K. Vánky, based on *Tilletia thlaspeos*, and ***Bauerago*** K. Vánky, based on *Ustilago abstrusa*. **New combinations** proposed: *Tothiella thlaspeos* (Beck) K. Vánky, based on *Tilletia thlaspeos* (type on *Thlaspi alpestre*, Austria), *Bauerago abstrusa* (Malençon) K. Vánky, based on *Ustilago abstrusa* (type on *Juncus gerardii*, France), *B. capensis* (Reess) K. Vánky, based on *Ustilago capensis* (type on *Juncus capensis*, South Africa), *B. cyperi-lucidi* (J. Walker) K. Vánky, based on *Ustilago cyperi-lucidi* (type on *Cyperus lucidus*, Australia), *B. gardneri* (McKenzie & K. Vánky) K. Vánky, based on *Ustilago gardneri* (type on *Cyperus ustulatus*, New Zealand), and *B. vuyckii* (Oudem. & Beij.) K. Vánky, based on *Ustilago vuyckii* (type on *Luzula campestris*, Netherlands).

Key words: smut fungi, classification, *Tothiella*, *Bauerago*.

INTRODUCTION

Based on the basidial type Tulasne & Tulasne (1847) divided smut fungi into two families, Ustilaginaceae and Tilletiaceae. Few changes have been made to this fossilised system of classification until recently. Data have accumulated which demonstrated on the one hand that smut fungi are not homogenous, and on the other hand that different microfungi are closely related to the smut fungi. The first surprising result, based on molecular biological data (5S rRNA analyses by Blanz & Gottschalk, 1984), and supported also by biochemical analyses (Prillinger et al., 1991) was the demonstration of the isolated position of some smut fungi, classified today into the Order Microbotryales R. Bauer & Oberw. Based on studies of nuclear LSU rDNA sequences, Begerow et al. (1997) placed the Microbotryales into the Class Uredinomycetes, together with the Order

Uredinales and other fungi. This confirmed the new classification of Bauer et al. (1997), elaborated on results obtained by ultrastructural studies. Thus, the Microbotryales are "blood-relatives" of the rust fungi despite in all respects looking like true smut fungi. For more details about the history of *Microbotryum* see Vánky, 1998a.

Based on ultrastructural studies (Bauer et al., 1997), confirmed and somewhat modified by molecular biological studies (Begerow et al., 1997), a new system for classification of the smut fungi and related taxa was elaborated (see also Bauer et al., in press). This system, completed by additions of the most recent data, can be summarised as follows. The number of known species within each genus is given within parentheses. The host families parasitised are also listed, together with Mo (for monocotyledonous hosts) and Di (for dicotyledonous hosts).

I. Cl. **USTILAGINOMYCETES** R. Bauer, Oberw. & Vánky

1. Subcl. **ENTORRHIZOMYCETIDAE** R. Bauer & Oberw.
    - Ord. **ENTORRHIZALES** R. Bauer & Oberw.
        - Fam. **Entorrhizaceae** R. Bauer & Oberw.
            - *Entorrhiza* C. Weber (9) Mo (Cyperaceae, Juncaceae)
2. Subcl. **USTILAGINOMYCETIDAE** Jülich, emend. R. Bauer & Oberw.
    1. Ord. **UROCYSTALES** R. Bauer & Oberw.
        1. Fam. **Melanotaeniaceae** Begerow, R. Bauer & Oberw.
            - *Exoteliospora* R. Bauer, Oberw. & Vánky (1) Pteridophyta (Osmundaceae) [in preparation]
            - *Melanotaenium* de Bary, s. str. (c. 10) Di (8 fam.)
        2. Fam. **Doassansiopsaceae** Begerow, R. Bauer & Oberw.
            - *Doassansiopsis* (Setch.) Dietel (9) Mo (3 fam.), Di (Nymphaceae)
        3. Fam. **Urocystaceae** Begerow, R. Bauer & Oberw.
            - *Mundkurella* Thirum. (4) Di (Araliaceae)
            - *Urocystis* Rabenh. (133) Mo (6 fam.), Di (15 fam.)
            - *Ustacystis* Zundel (1) Di (Rosaceae)
            - "*Melanotaenium*" *ari* (Cooke) Lagerh.
    2. Ord. **USTILAGINALES** G. P. Clinton, emend. R. Bauer & Oberw.
        1. Fam. **Mycosyringaceae** R. Bauer & Oberw.
            - *Mycosyrinx* Beck (3) Di (Vitaceae)
        2. Fam. **Glomosporiaceae** Cif., emend. Begerow, R. Bauer & Oberw.
            - *Glomosporium* Kochman (1) Di (Chenopodiaceae)
            - *Thecaphora* Fingerh. [incl. *Sorosporium*] (c. 50) Di (13 fam.)
            - *Tothiella* Vánky (1) Di (Brassicaceae)
        3. Fam. **Ustilaginaceae** Tul. & C. Tul., emend. R. Bauer & Oberw.
            - *Cintractia* Cornu (c. 30) Mo (Cyperaceae, Juncaceae)
            - *Clintamra* Cordas & Durán (1) Mo (Liliaceae)
            - *Dermatosorus* Sawada ex L. Ling (5) Mo (Cyperaceae)
            - *Farysia* Racib. (c. 20) Mo (Cyperaceae)
            - *Franzpetrakia* Thirum. & Pavgi, emend. L. Guo, Vánky & Mordue (2) Mo (Poaceae)
            - *Geminago* Vánky & R. Bauer (1) Di (Sterculiaceae)

Heterotolyposporium Vánky (2) Mo (Cyperaceae, Juncaceae)
Kuntzeomyces Henn. ex Sacc. & P. Syd. (1) Mo (Cyperaceae)
Macalpinomyces Langdon & Full., emend. Vánky (11) Mo (Poaceae)
Melanopsychium Beck (2) Di (Polygonaceae)
Moesziomyces Vánky (2) Mo (Poaceae, Eriocaulaceae)
Moreaua T. N. Liou & H. C. Cheng (c. 30) Mo (Cyperaceae)
Orphanomyces Savile (3) Mo (Cyperaceae)
Pericladium Pass. (4) Di (Tiliaceae, Piperaceae)
Schizonella Schröt. (5) Mo (Cyperaceae)
Sporisorium Ehrenb. (c. 150) Mo (Poaceae)
Tolyposporium Woronin ex Schröt. (c. 30) Mo (Cyperaceae, Juncaceae)
Trichocintractia M. Piepenbr. (1) Mo (Cyperaceae)
Ustilago (Pers.) Roussel (c. 240) Mo (Poaceae)
Websdanea Vánky (1) Mo (Restionaceae)
? *Testicularia* Klotzsch (1) Mo (Cyperaceae)
[? *Tranzscheliella* Lavrov (1) Mo (Poaceae)]
? *Uleiella* Schröt. (2) Di (Araucariaceae)
4. Fam. **Anthracoideaceae** Denchev
Anthracoidea Bref. (c. 80) Mo (Cyperaceae)
? *Planetella* Savile (1) Mo (Cyperaceae)
3. Subcl. **EXOBASIDIOMYCETIDAE** Jülich, emend. R. Bauer & Oberw.
1. Ord. **GEORGEFISCHERIALES** R. Bauer, Begerow & Oberw.
1. Fam. **Georgefischeriaceae** R. Bauer, Begerow & Oberw.
Georgefischeria Thirum. & Narash., emend. Gandhe (2) Di (Convolvulaceae)
Tolyposporella G. F. Atkinson (1-6?) Mo (Poaceae)
?*Jamesdicksonia* Thirum., Pavgi & Payak (1) Mo (Poaceae)
some "*Entyloma*" and "*Melanotaenium*" species on Poaceae and Cyperaceae
2. Fam. **Tilletiariaceae** R. T. Moore
Tilletiaria Bandoni & B. N. Johri
some "*Entyloma*" and "*Melanotaenium*" spp. on Poaceae and Cyperaceae
2. Ord. **TILLETIALES** Kreisel ex R. Bauer & Oberw.
Fam. **Tilletiaceae** Tul. & C. Tul., emend. R. Bauer & Oberw.
Conidiosporomyces Vánky (2) Mo (Poaceae)
Erratomyces M. Piepenbr. & R. Bauer (5) Di (Fabaceae)
Ingoldiomyces Vánky (1) Mo (Poaceae)
Neovossia Körn. s. str. (1) Mo (Poaceae)
Oberwinkleria Vánky & R. Bauer (1) Mo (Poaceae)
Tilletia Tul. & C. Tul. (c. 120) Mo (Poaceae)
3. Ord. **MICROSTROMATALES** R. Bauer & Oberw.
Fam. **Microstromataceae** Jülich
Microstroma Niessl (3) Di (Juglandaceae, Fagaceae)
4. Ord. **ENTYLOMATALES** R. Bauer & Oberw.
Fam. **Entylomataceae** R. Bauer & Oberw.
Entyloma de Bary, s. str. (c. 165) Di (Asteraceae 77, + 23 fam.)
5. Ord. **DOASSANSIALES** R. Bauer & Oberw.
1. Fam. **Doassansiaceae** (Azbukina & Karatygin) R. T. Moore, emend. R. Bauer & Oberw.
Burrillia Setch. (7) Mo (Alismataceae, Pontederiaceae) + Di (Gentianaceae)

Doassansia Cornu (19) Mo + Di
Doassinga Vánky, R. Bauer & Begerow (1) Di (Callitrichaceae)
Heterodoassansia Vánky (5) Mo + Di
Nannfeldtiomyces Vánky (2) Mo (Sparganiaceae)
Narasimhania Thirum. & Pavgi, emend. Vánky (1) Mo (Alismataceae)
Pseudodermatosorus Vánky (1) Mo (Alismataceae) [in preparation]
Pseudodoassansia (Setch.) Vánky (1) Mo (Alismataceae)
Tracya Syd. & P. Syd. (3) Di (Hydrocharitaceae, Lemnaceae)

2. Fam. **Rhamphosporaceae** R. Bauer & Oberw.
Rhamphospora D. D. Cunn. (1) Di (Nymphaeaceae)

3. Fam. **Melaniellaceae** R. Bauer, Vánky, Begerow & Oberw. [in preparation]
Melaniella R. Bauer, Vánky, Begerow & Oberw. (2) Pteridophyta (Selaginellaceae) [in preparation]

6. Ord. **EXOBASIDIALES** Henn., emend. R. Bauer & Oberw.

1. Fam. **Brachybasidiaceae** Gäum.
Brachybasidium Gäum. (?) Mo (Arecaceae)
Dicellomyces L. S. Olive (?) Mo (Poaceae, Cyperaceae)
Exobasidiellum Donk (?) Mo (Poaceae)
Kordyana Racib. (?) Mo (Commelinaceae)
Proliferobasidium J. L. Cunn. (?) Mo (Heliconiaceae)

2. Fam. **Exobasidiaceae** Henn.
Exobasidium Woronin (?) Di
Muribasidiospora Kamat & Rajendren (?) Di (Anacardiaceae, Ulmaceae)

3. Fam. **Cryptobasidiaceae** Malençon ex Donk
Botryoconis Syd. & P. Syd. (?) Di (Lauraceae)
Clinoconidium Pat. (?) Di (Lauraceae)
Coniodictyum Har. & Pat. (?) Di (Rhamnaceae)
Drepanoconis Schröt. & Henn. (?) Di (Lauraceae)
"*Sphacelotheca*" *cinnamomi* S. Hirata (1) Di (Lauraceae)
"*Ustilago*" *onumae* Shirai (1) Di (Lauraceae)

4. Fam. **Graphiolaceae** E. Fisch.
Graphiola A. Poit. (?) Mo (Arecaceae)
Stylina Syd. (?) Mo (Arecaceae)

II. Cl. **UREDINOMYCETES** (author?)

1. Ord. **MICROBOTRYALES** R. Bauer & Oberw.

1. Fam. **Microbotryaceae** R. T. Moore, emend. R. Bauer & Oberw.
Liroa Cif. (1) Di (Polygonaceae)
Microbotryum Lév., emend. G. Deml & Oberw., emend. G. Deml & Prillinger, emend. Vánky (75) Di (9 fam.)
Sphacelotheca de Bary (5) Di (Polygonaceae)
Zundeliomyces Vánky (1) Di (Polygonaceae)

2. Fam. **Ustilentylomataceae** R. Bauer & Oberw.
Aurantiosporium M. Piepenbr., Vánky & Oberw. (2) Mo (Cyperaceae)
Fulvisporium Vánky (1) Mo (Poaceae)
Ustilentyloma Savile (3) Mo (Poaceae)
?*Bauerago* Vánky (5) Mo (Cyperaceae, Juncaceae)

2. Ord. **UREDINALES**
etc.

Analysing many years of observations, and the most recent results concerning classification of the smut fungi, it became clear that the true *Ustilago* species are restricted to monocotyledonous host plants only, specifically to Poaceae. It also became obvious that all "*Ustilago*" species of dicotyledonous host plants possessing violet-coloured spores belong to the genus *Microbotryum* (Vánky, 1998a). The question, which automatically rises, is to which genera do the *Ustilago* species of dicotyledonous host plants, which do not possess violet-coloured spores, belong? These are either not smut fungi, or they belong to already known or to still undescribed genera. For example, it turned out that *Ustilago(?) senecionis* Dietel & Neger (on *Senecio glaber* Less., Asteraceae) is a myxomycete (Vánky, 1998b), *U. gunnerae* G. P. Clinton (nom. nud.; on *Gunnera magellanica* Lam., Haloragaceae) is a *Melanotaenium* species (*M. gunnerae* G. P. Clinton ex K. Vánky & R. Bauer), *U. speculariae* J. A. Stevens (on *Specularia perfoliata* (L.) DC., Campanulaceae) belongs to a still undescribed genus. This is also the case with *Ustilago thlaspeos*, for which a new genus is proposed:

Tothiella K. Vánky, **gen. nov.**

Sporeae singulae, pigmentiferae (brunneae), sine ullo colore violaceo, in seminibus plantarum dicotyledonearum productae. Massa sporarum pulverea. Peridium circa soros, cellulae steriles et columellae inter sporas, hae omnes absentes. Interactio inter plantas nutrientes et parasitas per hyphas intracellulares. Septa mature sine poris. Germinatio sporarum cum hyphis ramosis. Anamorphae forte praesentes. Typus generis *T. thlaspeos*.

Spores single, pigmented (brown), lacking violet tint, produced in the seeds of dicotyledonous host plants. Spore mass powdery. Peridium around the sori, sterile cells and columellae between the spores are lacking. Host–parasite interaction by intracellular hyphae. Mature septa are poreless. Spore germination results in ramifying hyphae. Anamorph may be present. Typus generis:

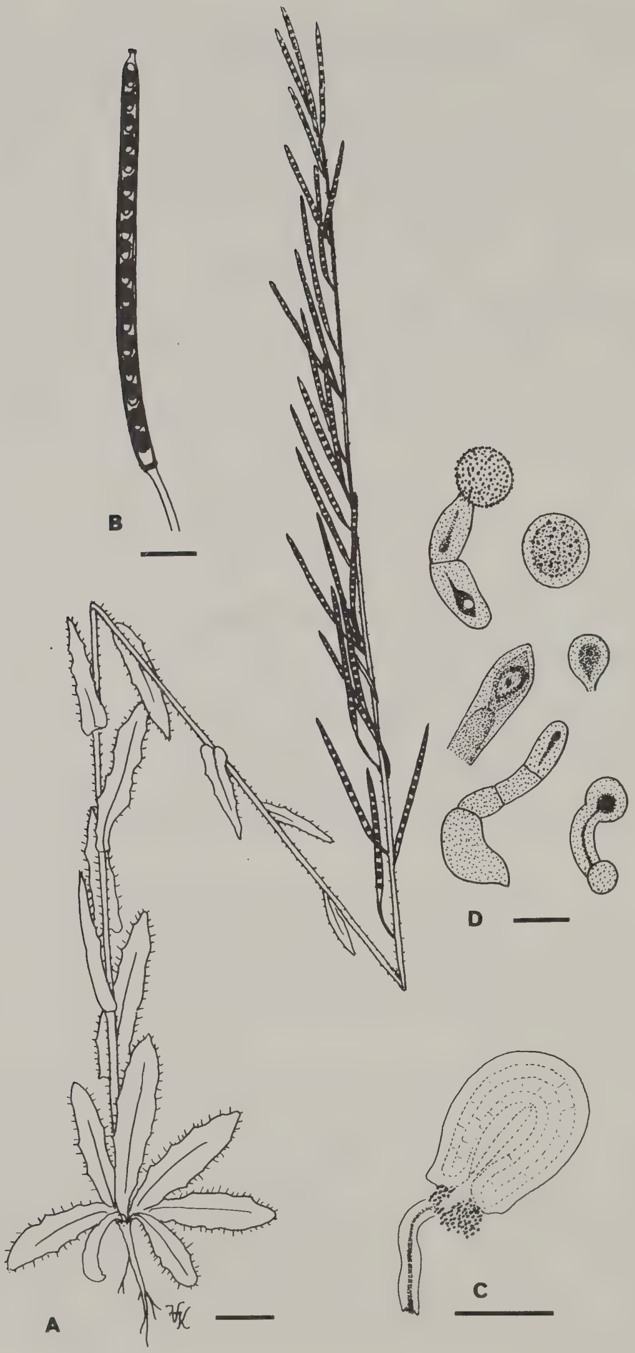
Tothiella thlaspeos (Beck) K. Vánky, **comb. nov.**

Basionym: *Tilletia thlaspeos* Beck, Verh. K. K. Zool.-Bot. Ges. Wien 35:362, 1886. – *Ustilago thlaspeos* (Beck) Lagerheim, in Sydow, Ust. No. 118, 1897. – *Bauhinus thlaspeos* (Beck) Denchev, Mycotaxon 65:424, 1997. – Type on *Thlaspi alpestre* L., Austria, Burgenland, near Redlschlag [Újvörösvágás], coll. V. Borbás (HUV 4776!).

Ustilago seminum Juel, Öfvers. Förh. Kongl. Svenska Vetensk.-Akad. 51:491, 1894. – Type on *Arabis petraea* Lam. (= *Cardaminopsis petraea* (L.) Hiitonen, Norway, Oppland, Sulheim near Lom, 17.VI.1894, O. Juel; isotypes in Eriksson, Fgi. paras. scand. No. 454 (HUV 4787!).

Ustilago arabidis-alpinae Liro, Ann. Acad. Sci. Fenn., Ser. A, 42(1):507, 1938 (illeg. name, no Latin dg.); Mycotheca fennica. Die Etiketten. No. 301-600, p. 108, 1939. – Type on *Arabis alpina* L., Finland, Lapponia enontekiensis, Kilpisjärvi, the E. slope of Saana Mt., alt. 750 m, 28.VII.1936, J. I. Liro (H).

Ustilago cardamines Liro, Ann. Acad. Sci. Fenn., Ser. A, 42(1):507, 1938 (illeg. name, no Latin dg.); Mycotheca fennica. Die Etiketten. No. 301-600,



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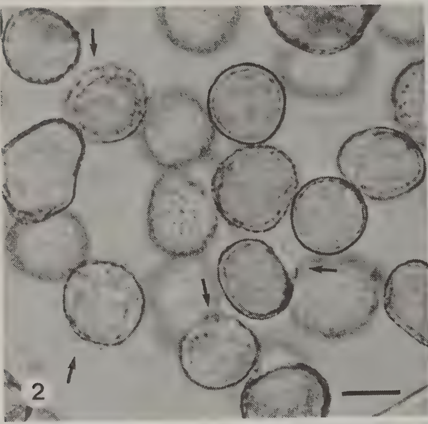
p. 108, 1939. – Lectotype on *Cardamine bellidifolia* L., Finland, Lapponia enontekiensis, (designated by Lindeberg, Symb. Bot. Upsal. 16(2):140, 1959) Meekonjauri, Perfevanghi, 6.VIII.1936, J. I. Liro & H. Roivainen; isolectotypes in Mycoth. fenn. 792 (HUV 7747!).

Sori (Fig. 1) in the seeds, beginning to develop at the funiculus, gradually replacing the whole seed with a light brown, dusty, rather inconspicuous spore mass of single spores which is liberated when the siliquae open. Infection systemic. The spores are differentiated within the septate mycelia, one spore in each cell (Vánky, 1970:326; Fig. 1 D). *Spores* (Figs. 2, 3) globose, subglobose, ovoid to irregular, 10–16 x 11–18 µm, yellow to light yellowish-brown lacking violet tint; wall even, c. 1 µm thick, with moderately densely situated, irregular, 0.5–1 µm high warts, the warts are denser, more elongated (1.5–2.5 µm high) and often acute on a restricted area of the spores; in SEM fine, dense verruculae lie between the irregular warts, tubercles or spines. *Spore germination* results in ramifying hyphae (Fig. 4). [Juel, 1894:493, records ramifying, septate hyphae, but his illustrations show only retraction septae]. *Host–parasite interaction* by intracellular hyphae (Fig. 6). Mature *septa* are poreless (Fig. 7). *Anamorph* (*Rhombiella cardamines* Liro) is present in the anthers.

On Cruciferae: *Alyssum reiseri* Vel., *Arabis alpina* L., *Ar. corymbiflora* Vest., *Ar. hirsuta* (L.) Scop., *Cardamine bellidifolia* L., *Cardaminopsis petraea* (L.) Hiitonen, *Draba aizoides* L., *D. alpina* L., *D. incana* L., *Erysimum diffusum* Ehrh., *E. weltschewii* Urum., *Thlaspi alpestre* L., *T. alpinum* Crantz; Europe.

The morphological and ultrastructural characters of *Tothiella thlaspeos* are identical with those of *Thecaphora saponariae* (Rudolphi) K. Vánky (*Sorosporium saponariae* Rudolphi) except for the absence of spore balls in *Tothiella thlaspeos*. Its place in the new system is in the Glomosporiaceae beside *Glomosporium* and *Thecaphora*, two genera characterised by spore balls. It seems that the presence or absence of spore balls is not as strong a character of relationship between different groups of smut fungi as was thought earlier. For example, *Doassinga* K. Vánky, R. Bauer & Begerow is a genus with single spores belonging to the Doassansiales R. Bauer & Oberw., together with seven additional genera, all characterised by spore balls. It is interesting to mention that Juel (1894:493), in connection with *Ustilago seminum* Juel (= *Tothiella thlaspeos*), based on morphological characters of the spores and the similar type of spore germination wrote: "Ich glaube daher, dass *U. seminum* und *Sorosporium saponariae* nahe verwandte Arten sind" [I think, that *U. seminum* and *S. saponariae* are closely related species].

Fig. 1. Sori of *Tothiella thlaspeos* (Beck) K. Vánky on *Arabis hirsuta* (L.) Scop. (Vánky, Ust. exs. No. 96, as *Ustilago thlaspeos*). **A**. Habit (Bar = 1 cm). **B**. Enlarged, opened siliqua with sori in the seeds (Bar = 3 mm). **C**. A young seed with maturing spores in the funiculus–hilum region (Bar = 0.5 mm). **D**. Sporogenous hyphae with spore initials of different ages (Bar = 10 µm).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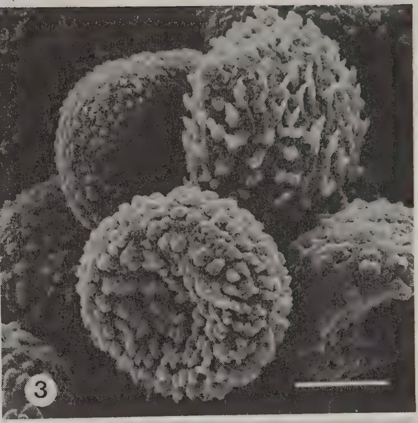
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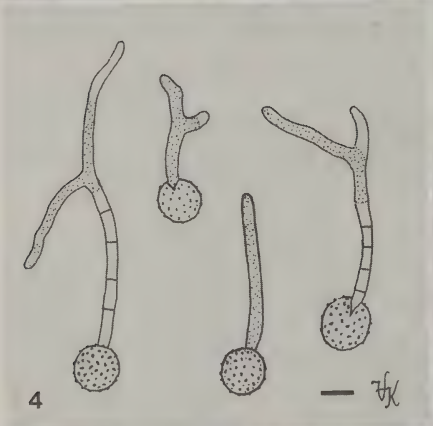
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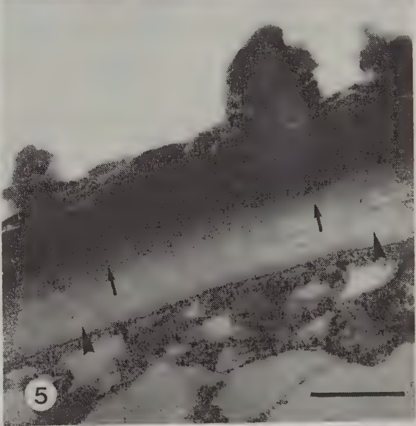
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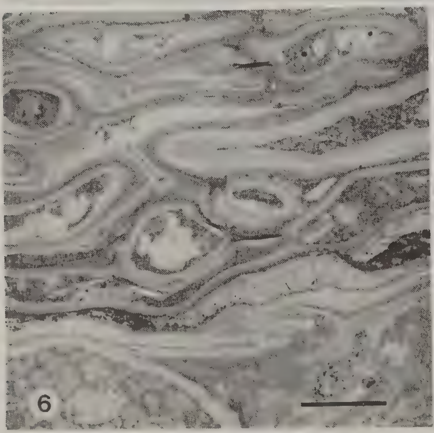
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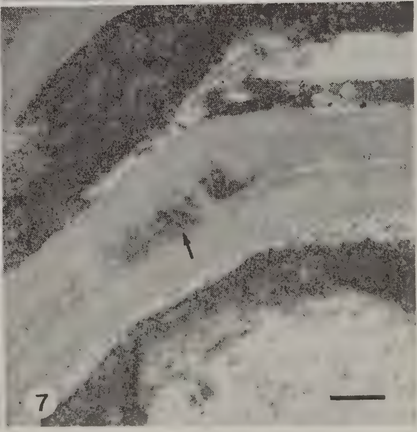
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Tothiella differs from *Microbotryum*, also on dicotyledonous host plants, i.a., in the morphology (including colour) of the spores and in the host–parasite interaction. In *Microbotryum* species the spores are more regular, have a more or less expressed violet colour, and the parasitic hyphae are intercellular.

Etymology: *Tothiella* is named in the honour of Dr. Sándor Tóth on the occasion of his 80th birthday. Dr. S. Tóth is the most outstanding Hungarian contemporary mycologist, with great national and international merits in the field of microfungi. He is an excellent, unselfish human and friend who, despite his handicaps, is ready to help everybody.

Since true *Ustilago* species are restricted to host plants in the Poaceae, and knowing the importance of the colour of the spores (chemotaxonomy!), it is obvious that "*Ustilago*" species of Cyperaceae and Juncaceae possessing yellow or yellowish-red spore masses and spores must have a different systematic position. Smut fungi possessing yellow or yellowish-red spore masses and spores and characterised by more or less permanent spore balls are members of the genera *Aurantiosporium* M. Piepenbr., K. Vánky & Oberw., and *Fulvisporium* K. Vánky. In the new system they are placed in the family of Ustilentylomataceae. For the single-spored, yellow- or yellowish-red coloured "*Ustilago*" species of Cyperaceae and Juncaceae the erection of a separate genus is necessary, for which I am proposing the name of:

Figs. 2–7. *Tothiella thlaspeos* (Beck) K. Vánky on *Arabis hirsuta* (L.) Scop. (Vánky, Ust. exs. No. 96).

Figs. 2, 3. Spores in LM and in SEM. Note the more intense ornamentation in one side of the spores (arrows). Bars = 10 µm and 5 µm, respectively.

Fig. 4. Germinating spores (on water, at room temp., after 4 days) giving rise to branched hyphae with retraction septa. Bar = 10 µm.

Fig. 5. Transverse section of a spore wall in TEM, showing an electron-transparent endospore (arrow heads), a thin, medium electron-dense middle layer (arrows), and an electron-dense exospore with ornamentation, having a lamellate structure (Photo R. Bauer). Bar = 0.5 µm.

Fig. 6. Host–parasite interaction, realised by intracellular hyphae. The upper 2/3-rd of the picture shows a part of a host cell filled with hyphae (Photo R. Bauer). Bar = 2 µm.

Fig. 7. Mature septal wall is three-layered, with an interrupted middle lamella (arrow). Pore is lacking (Photo R. Bauer). Bar = 0.2 µm.

Bauerago K. Vánky, **gen. nov.**

Sporae singulares, flavae usque rubiginosae pigmentiferae, sine ullo colore violaceo, in seminibus plantarum monocotyledonearum (Cyperaceae, Juncaceae) productae. Massa sporarum pulverea. Peridium circa soros, cellulae steriles et columellae inter sporas hae omnes absentes. Germinatio sporarum cum phragmobasidiis. Interactio inter plantas nutrientes et parasitas per hyphas intercellulares. Anamorphae forte praesentes. Typus generis *B. abstrusa.*

Spores single, yellow to rusty-brown pigmented, lacking violet tint, produced in the seeds of monocotyledonous host plants (Cyperaceae, Juncaceae). Spore mass powdery. Peridium around the sori, sterile cells and columellae between the spores are lacking. Spore germination results in phragmobasidia. Host–parasite interaction by intercellular hyphae. Anamorph may be present. Typus generis:

Bauerago abstrusa (Malençon) K. Vánky, **comb. nov.**

Basionym: *Ustilago abstrusa* Malençon, Bull. Soc. Mycol. France 45:256, 1929. – *Cintractia abstrusa* (Malençon) Ciferri, Ann. Mycol. 29:56, 1931. – Type on *Juncus gerardii* Loisel., France, Manche Dépt., Gatteville near Cherbourg, 19.IX.1926, G. Malençon (MPU).

Sori (Fig. 8) in ovaries filling the capsules with golden- to rusty-brown, dusty spore masses. Infection systemic, not causing conspicuous deformation. *Spores* (Figs. 10, 11) globose, ovoid, often slightly subpolyhedrally irregular, 13–17(–18) x 14–20(–22) µm, from subhyaline to lemon- or orange-yellow, finely reticulate, 7–12 meshes per spore diameter, muri 1–1.5 µm high, in optical median view blunt. Host–parasite interaction by intercellular hyphae.

On Juncaceae: *Juncus balticus* Willd., *J. balticus x filiformis, J. gerardii* Loisel., *J. gregiflorus* L. Johnson; Europe, New Zealand.

Other species in the genus are:

Bauerago capensis (Reess) K. Vánky, **comb. nov.**

Basionym: *Ustilago capensis* Reess, in Buchenau, Abh. Naturwiss. Vereins Bremen 4:486, 1875; Sitzungsber. Phys.-Med. Soc. Erlangen 7:70, 1875. – Type on *Juncus capensis* Thunb. var. *longifolius* Eckl. & Zeyher, South Africa, Cape of Good Hope, mountains at Cape Town, alt. 2000 feet, coll. M. Reess.

Sori in the seeds. *Spores* (judged from Reess' illustrations) deeply reticulate, c. 5 meshes per spore diameter. No specimen of this species could be located.

On Juncaceae: *Juncus capensis* Thunb., *J. capensis* Thunb. var. *longifolius* Eckl. & Zeyher, *J. lomatophyllus* Sprengel; South Africa.

Bauerago cyperi-lucidi (J. Walker) K. Vánky, **comb. nov.**

Basionym: *Ustilago cyperi-lucidi* J. Walker, Proc. Linn. Soc. New South Wales 96:99, 1971. – Type on *Cyperus lucidus* R. Br., Australia, New South Wales, W. of Pambula, Wyndham, 11.VII.1969, J. Hindle (DAR 17587/a; it. HUV 14062!).

Similar to *B. gardneri,* but the spores of *B. cyperi-lucidi* are (12–)16–20(–24) x (15–)17–24(–26) µm, the spore wall is 2.5–4 µm thick, the muri 1–2 µm high.

On Cyperaceae: *Cyperus lucidus* R. Br.; Australia.

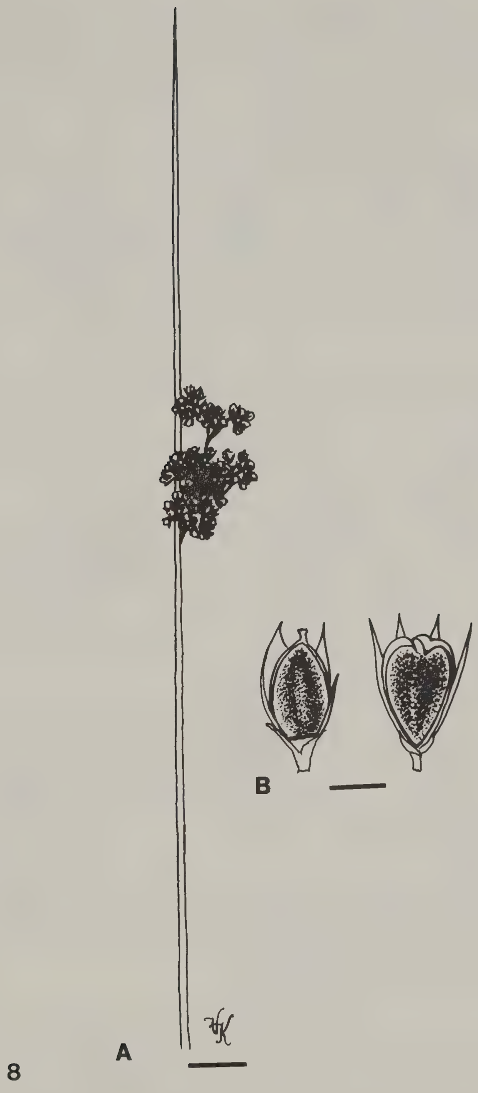

Fig. 8. Sori of *Bauerago abstrusa* (Malençon) K. Vánky in the capsules of *Juncus gregiflorus* L. Johnson. **A**. Habit. **B**. Two longitudinally sectioned capsules filled with spore masses (Vánky, Ust. exs. No. 1002).
Bars = 1 cm and 1 mm, respectively.

Bauerago gardneri (McKenzie & K. Vánky) K. Vánky, **comb. nov.**
Basionym: *Ustilago gardneri* McKenzie & K. Vánky, in Vánky, Mycotaxon 41:486, 1991. – Type on *Cyperus ustulatus* A. Rich. f. *grandispiculosus* Kük. ex Carse, New Zealand, North Island, Auckland, Western Springs Lake, 17.II.1989, R. O. Gardner (PDD 57462, ht; BPI, it; HUV 14850, it).

Sori in swollen nutlets, filled by a light ochraceous-yellow, powdery mass of spores exposed upon the rupture of the pericarp. *Spores* subglobose, ovoid to slightly subpolyhedrally irregular, 13–18(–20) x 15–20(–21) µm, lemon-yellow; wall 2–2.5 µm thick, very finely and minutely reticulate, sometimes incompletely, 10–15 meshes per spore diameter; including the reticulum, muri 0.8–1 µm high. Infection systemic, all nutlets in the inflorescence being affected.

On Cyperaceae: *Cyperus ustulatus* A. Rich.; New Zealand.

Bauerago vuyckii (Oudem. & Beij.) K. Vánky, **comb. nov.**
Basionym: *Ustilago vuyckii* Oudem. & Beij., in Oudemans, Versl. Zittingen Wis.-Naturk. Afd. 3:(55), 1895. – *Cintractia vuyckii* (Oudem. & Beij.) Ciferri, Ann. Mycol. 29:72, 1931. – Type on *Luzula campestris* (L.) DC., Netherlands, near Voorschoten, 22.V.1894, L. Vuyck (L!, LE!).

Sori in ovaries filling the capsules with an ochraceous-yellow, dusty spore mass. Infection systemic; infected plants may be slightly deformed. *Spores* globose to ellipsoidal, 13–20 x 16–21 µm, pale yellow to light yellowish-brown; wall 2–3.5 µm thick, deeply reticulate, 5–8 meshes per spore diameter, meshes 2–3 µm wide, muri 1.5–2.5(–3) µm high, in optical median view acute or subacute. *Anamorph* was reported in the anthers.

On Juncaceae: *Luzula campestris* (L.) DC., *L. glabrata* Desv., *L. multiflora* (Retz.) Lej. subsp. *multiflora*, and subsp. *frigida* (Buchenau) V. Krecz. (*L. frigida* (Buchenau) Sam.), *L. novae-cambriae* Gandoger, *L. parviflora* (Ehrh.) Desv., *L. pilosa* (L.) Willd. (*L. vernalis* (Reichard) DC.), *Luzula sibirica* V. Krecz.; Europe, Asia, North America, Australia.

Etymology: *Bauer-ago*, derived from *Bauer* and Ustil*ago*. Dr. Robert Bauer, German mycologist from Tübingen, one of the world's most outstanding specialists in fungal ultrastructure, who, i.a., elaborated the new system for the systematics of the smut fungi and related taxa.

Knowing the characters of the 62 genera of "classical" smut fungi and their new classificatory system, *Bauerago*, with all probability, belongs to the Ustilentylomataceae within the Microbotryales, especially if it will turn out that the septa have simple pores, similar to those in *Fulvisporium* and *Aurantiosporium* (comp. Vánky et al., 1997, and Vánky, 1999, in press). The presence of intercellular hyphae, seen in LM, supports this assumption.

In sori of *Bauerago abstrusa*, on the surface of young ovaries, successive spore building in the cells of sporogenous hyphae, and also clamp connections between these cells could be observed (Fig. 9), very similar to those reported by Liro (1924:517-522) and Seyfert (1927) for *Ustilago vuyckii* (= *Bauerago v.*).

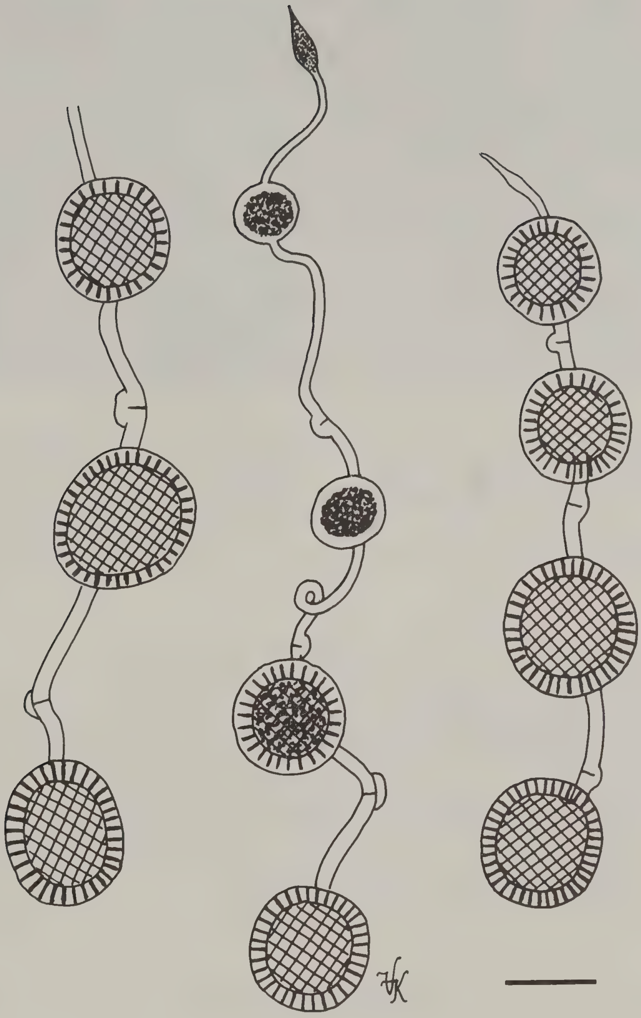

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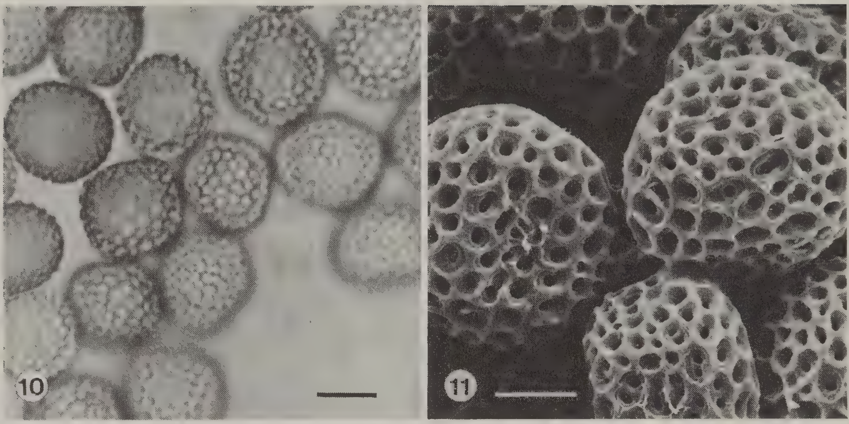
Fig. 9. Spore formation in the hyphae of *Bauerago abstrusa* (Malençon) K. Vánky. Freshly collected, young sori from *Juncus gregiflorus* were fixed in glutaraldehyde, squashed and coloured with cotton blue in lactophenol. Between cells of sporogenous hyphae, clamp connections can be seen. Spore ornamentation schematical (Vánky, Ust. exs. No. 1002). Bar = 10 µm.



Figs. 10, 11. Spores of *Bauerago abstrusa* (Malençon) K. Vánky from *Juncus gregiflorus* in LM and in SEM (New Zealand, North Canterbury, Horseshoe Lake, 30.I.1990, E. H. C. McKenzie & K. Vánky, HUV 14972).
Bars = 10 µm and 5 µm, respectively.

Key to the *Bauerago* species.

1. On Juncaceae 2
– On Cyperaceae (*Cyperus*) 4
2. On *Luzula*. Spores deeply reticulate (muri 1.5–2.5(–3) µm high), 5–8 meshes per spore diam. .... *B. vuyckii*
– On *Juncus* 3
3. Spores shallowly reticulate, 7–12 meshes per spore diam. .... *B. abstrusa*
– Spores deeply reticulate, c. 5 meshes per spore diam. .... *B. capensis*
4. Spore wall 2.5–4 µm thick. Muri 1–2 µm high .... *B. cyperi-lucidi*
– Spore wall 2–2.5 µm thick. Muri 0.8–1 µm high .... *B. gardneri*

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MYCOTAXON

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SEPTORIA BIRGITAE SP. NOV., A NEW PATHOGEN CAUSING LEAF SPOTS ON LACTUCA SATIVA

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SUMMARY

Septoria birgitae Bedlan sp. nov. collected on *Lactuca sativa* L. differs from other species of *Septoria* on *Lactuca* and *Mulgedium* in the diameter of the pycnidia, length, width and number of septa of the conidia.

KEY WORDS

Septoria birgitae Bedlan sp. nov.; *Lactuca sativa* L.; symptoms; biology.

In the cool and mostly rainy year 1996 the leaves of lettuce (*Lactuca sativa* L.) frequently showed brown spots. After carrying out microscopic tests we found out that it was a fungus of the genus *Septoria*.

In absence of suitable hosts, however, the pathogen can only survive for a short time. For the related species *Septoria lactucae* Pass. it is said that the fungus is transported over long distances together with the seed, whereas from plant to plant it is transferred by water drops (rain, irrigation), animals, tools and harvesting (CHUPP & CHERF, 1960). This should also apply to *Septoria birgitae*.

The first samples of *Septoria birgitae* were taken from biological farming in 1996, from the lettuce varieties „Kermit", „Libusa" and „Florial". Finally lots of lettuce varieties all over Austria were found to be infested. *Septoria birgitae* has also been found in lettuce quite frequently in Germany (KOFOET, 1996).

First symptoms are small, yellowish areas on the exterior leaves of lettuce. These may very soon turn brown, and finally spread over the whole leaf. Even young plants may be infested. On the spots, which are angular, round and sometimes with visible margin, the numerous pycnidia of the fungus can be seen with the naked eye.

The pycnidia are scattered over the spots and sometimes grouped together. The pycnidia are dark brown and blackish and give off the thready hyaline conidia, typical of *Septoria,* via the ostiolum. In humid or wet conditions the disease can occur very heavily. The conidia of *Septoria spp.* are dispersed through water drops during rain or irrigation. BOND (1941) described this form of dispersion for *Septoria lactucae* Pass.

The older leaves are usually the first to be infested. If the plants are badly infested, peduncles and flower organs, and thus also the seeds, can be infected during seed production.
This fungus should also survive in the seeds and remainders of the lettuce like *Septoria lactucae* Pass. (SMITH, 1961).

Table: *Septoria* species on *Lactuca* and *Mulgedium*

Species	Pycnidia Diameter in µ	Pycnidia, Diameter average in µ	Length of conidia: min.-max. Average in µ	Width of conidia: min.-max. Average in µ	Number of Septa
S. lactucae Pass., Typus	51,71-136,25	83,00 90[3])	13,65-39,49 22,4 [1])	1,04-2,42 1,71 [1])	0
S. lactucina Lobik	82,3-125	103,70	39,5-52,6 46,10	3-3,3 3,2	1-3
S. lactucicola E. & M.	42,16-82,84	66,26	18,92-42,82 30,80[2])	0,99-2,18 1,58	1-3
S. schembelii Melnik, Typus	25,78-82,19	58,70	16,94-55,50 35,58	1-2,62 1,67	0
S. ludoviciana Ell. & Ev.	75-80	77,50	15-25 20,00	2	0
S. fernandezii Unamuno	112	-	26-30,5[3]) 28,25	1,6-2[3]) 1,8	1
S. sikangensis Petrak	60-150	105,00	20-52 36,00	2-3 2,5	3-6
S. unicolor Wint.	very small[3])	very small [3])	26-32 29,00	1	0 -
S. sleumeri Petrak	60-80	70	15-42 28,5	1-1,5 1,25	0
S. mulgedii Thüm.	92,54-201,40	167	26-28[4])	4[4])	1
S. birgitae Bedlan (Wallern/Bgld).	80,57-194,33	135,25	18,92-39,03 28,87	1,28-2,30 1,81	1-3
S. birgitae Bedlan (Harthausen, FRG)	85,96-195,75	139,72	16,25-36,73 27,48	1,51-2,85 2,19	1-3
S. birgitae Bedlan (Lustadt,FRG)	90,26-169,93	132,14	19,60-36,89 27,88	1,65-3,08 2,28	1-3

[1]) according to the original description: 25-30 µ long and 1,7-2 µ wide; all other data based on personal testings

[2]) according to the original description 25-30µ long; all other data based on personal testings

[3]) according to the original description

[4]) according to the original description, all other data based on personal testings

Septoria birgitae **BEDLAN sp. nov.**

Maculae dispersae, orbiculares vel ellipticae, saepe irregulares, in foliis, primo exterioribus, saepe in marginibus. Pycnidia, hypophylla, irregulariter dispersa, subepidermata, globosa vel ellipsoidea, 80,57 - 195,75 µ diam., ostiolo planum, 22 - 45 µ (∅ 33 µ) in diam. Septa pycnidiorum inter 5 et 20 µ lata, saepissime inter 9 et 13 µ.

Conidia filiformia, hyalina, recta vel curvulata vel falcata, utrinque obtusiuscula, ± attenuata, 1-3-septata, 18,92 - 39,03 µ x 1,28 - 2,30 µ.

Ad folia viva *Lactucae sativae* L. var. *capitatae* L. (var. „Sander") in Wallern/Burgenland, Austria, 25.7.1996.

A speciebus aliis in generibus *Lactuca* et *Mulgedio* vigentibus differt imprimis pycnidiorum diametro 80 - 200 µ, saepissime 132 - 139 µ longo, septisque conidiorum 3.

The type specimen has been deposited in the Museum of Natural History, Vienna: *Septoria birgitae* Bedlan, collected on live leaves of *Lactuca sativa* L. var. *capitata* L., variety „Sander", in Wallern/Bgld., Austria, on 25.7.1996

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TITAEA FORMOSA ANAM. SP. NOV., A NEW FUNGICOLOUS HYPHOMYCETE FROM SPAIN

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Abstract

Titaea formosa anam. sp. nov., a hyphomycete isolated from an apothecium of *Dasyscyphus fuscenses* is described and illustrated. The species is characterized by hyaline, complex, somewhat bird-like, bilaterally symmetrical conidia consisting of one clavate basal cell, two lageniform upper lateral elements and three globose cells, one apical and two sublateral hanging from the lageniform branches, being the apical cell larger than the two sublateral ones. Notes on closely similar species are given, and a dichotomous key to the seven known species of the genus is provided.

Keywords: *Titaea*, *Dasyscyphus*, Hyphomycetes, fungicolous fungi, Spain.

Introduction

During a survey of lignicolous and foliicolous ascomycetes in Sierra de las Villuercas, Cáceres (Spain), one of us (F. A.) collected dead leaves of *Quercus suber* L. colonized by a discomycete which was identified as *Dasyscyphus fuscenses* (Pers.) Gray (Leotiaceae, Leotiales). Attempts to isolate the ascomycete from the ascomatal tissues resulted in the recovery

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of a second fungus, morphologically different to the ascomycete culture derived from ascospores. After several weeks' incubation, the fungal colonist developed conidia whose morphology resembled that of some species of *Titaea* Sacc., such as *T. hemileiae* Hansf., although with significant differences. We thus describe this fungus as a new species of *Titaea*. The new taxon is compared in detail with *T. hemileiae*, the most similar species within the genus, and a key to the species of *Titaea* is included.

Materials and Methods

The fungus was isolated from pieces of an apothecium of *Dasyscyphus fuscenses*, plated on cornmeal agar (CMA) amended with antibiotics (streptomycin and oxytetracyclin, 50 mg/l each). Sporulation of the culture was evident after 21 days growth at 22°C under fluorescent light on a 12 hour light/dark cycle. In the descriptions, capitalized color names are from Kornerup & Wanscher (1978). Potato dextrose agar (PDA), oatmeal agar (OMA) and cornmeal agar (CMA) were prepared media from Difco Laboratories. Microphotographs were taken with a Leitz Diaplan microscope equipped with differential interference contrast optics.

Two herbarium specimens of *Titaea hemileiae* were examined for comparison with our species: IMI 5007, holotype, on sori of the tropical rust *Hemileia* sp. formed on leaves of *Alafia clusioides*, Entebbe Road, Uganda, leg. C. G. Hansford; and IMI 49981 (b), on sori of *Hemileia vastatrix* on *Coffea arabica*, Nigeria, 19 June 1952, leg. R. N. Chapman-Jones. Microphotographs were taken from the latter specimen, due to its better preservation.

Description of the species

Titaea formosa Peláez, R.F. Castañeda et Arenal *anam. sp. nov.*

Ad fungus conidiales, Hyphomycetes pertinens. Hyphae septatae, ramosae, laeves, interdum moniliformes, hyalinae, 1.5-3.0 µm diam. Sporodochia rudimentaria, superficialia, alba, 30-70 µm diam., ex conidiophoris compactis vel laxis aggregatis, conspicuis, profuse ramosis, raro simplicibus, septatis, erectis vel prostratis, 40-60 x 6-7 µm, moniliformibus, hyalinis, laevibus, composita. Cellulae conidiogenae monoblasticae et polyblasticae, doliiformes usque subcylindricae, 5-12 x 3-4 µm, terminales et intercalares, determinatae vel interdum cum proliferationibus sympodialibus et clare denticulatae, laeves, hyalinae. Conidia

blastica, hyalina, levia, aequilateralia, plus minusve avis-similia, ex cellulis 4 formarum composita: (a) cellula basalis obconica-truncata usque ad leviter clavata-truncata, 6-9 x 3.0-3.5 µm, 1.0-1.5 µm lata infra, levia, interdum guttulata, schizolytice secedentia; (b) 2 brachiis lateralibus, divergentibus, non-septatis, interdum septatis ad basem, lageniformibus usque anguste pyriformibus, (7-)9-17 x 3-4 µm, 1.0-1.5 µm latis ad apicem, ex cellula basalis oriundis; (c) 2 cellulae globosae, 4.5-7.0 µm diam., interdum guttulatae, in parte inferiore brachiis lateralibus circa basim, portatae; (d) cellula apicalis globosa, 6-11 µm diam. Teleomorphosis ignota.
TYPUS: in ascomatis ex *Dasyscyphus fuscenses* (Pers.) Gray in foliis emortuis *Quercus suberis* L., Sierra de las Villuercas, Cáceres (Spain), 19 May 1996, leg. F. Arenal. Cultura sicca (holotypus) in Herbarium MA-Fungi (MA-37927). Isotypus in IMI (IMI 378711).

Conidial fungi, Hyphomycetes. Hyphae septate, branched, smooth-walled, sometimes moniliform, hyaline, 1.5-3.0 µm diam. Sporodochia rudimentary, superficial, white, 30-70 µm diam., conidiophores loosely to compactly united, conspicuous, usually profusely branched, rarely simple, septate, erect or prostrate, 40-60 x 6-7 µm, moniliform, hyaline, smooth-walled. Conidiogenous cells monoblastic and polyblastic, doliiform to subcylindrical, 5-12 x 3-4 µm, terminal and intercalary, determinate or sometimes with sympodial proliferations and truncate denticles, concurrently proliferating, smooth-walled, hyaline. Conidia blastic, seceding schizolytically, hyaline, smooth-walled, bird-like, bilaterally symmetrical, complex, composed of 4 kinds of elements: (a) one basal cell, obconic-truncate to clavate-truncate, 6-9 x 3.0-3.5 µm, 1.0-1.5 µm wide below, sometimes guttulate, (b) two lateral branches widely divergent, usually aseptate, occasionally developing one septum near the base, lageniform to narrow pyriform, (7-)9-17 x 3-4 µm, 1.0-1.5 µm wide at the apex, arising subapically from the basal cell; (c) two lower lateral cells, globose, 4.5-7.0 µm diam., sometimes guttulate, arising near the base from the lower side of the upper lateral branches; (d) one apical cell, globose, 6-11 µm diam. Teleomorph unknown.

ETYMOLOGY: From the Latin *formosa* = beautiful, in reference to the appearance of the conidia.
HABITAT: in apothecium of *Dasyscyphus fuscenses* (Pers.) Gray growing on dead *Quercus suber* L. leaves in forest litter.
SPECIMEN EXAMINED: Sierra de las Villuercas, Cáceres (Spain), 19 May 1996, leg. F. Arenal. Dried culture (holotype) in Herbarium MA-Fungi (MA-37927). Isotype: IMI 378711.

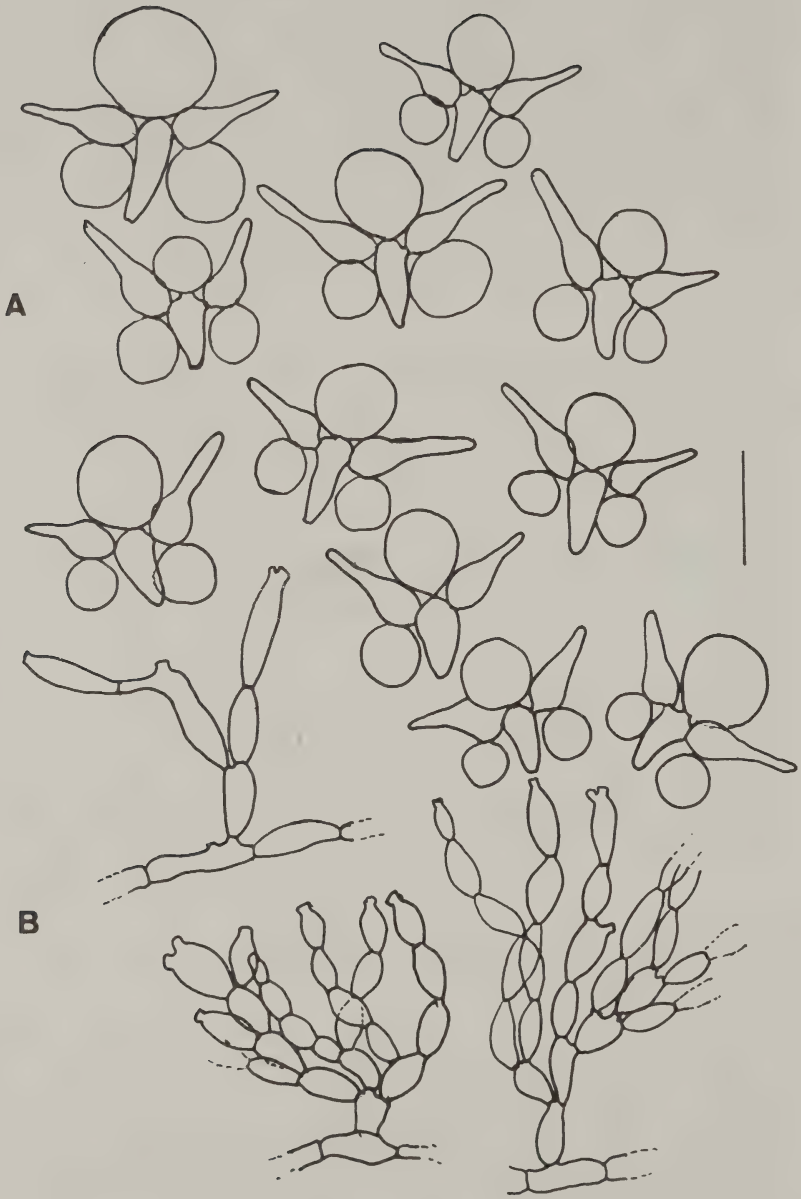


Fig 1. *Titaea formosa* sp. nov. A. Conidia. B. Conidiophores and conidiogenous cells. Bar: 10 µm.

CULTURAL CHARACTERS: Colonies on PDA relatively slow-growing, attaining 21-25 mm diam. in 5 weeks at 22°C, 12 hr photoperiod; growing slightly more rapidly at 18°C (24-35 mm) and slower at 28°C (16-23 mm). Margin uneven, appressed. Colony raised at the center, aerial mycelium crustose, irregularly wrinkled (cerebriform), radially sulcate toward the margins, greyish orange, reddish brown, brownish orange (Caramel, Sahara), greyish brown, brownish beige or light brown (Camel, Sunburn, Cinnamon). Reverse greyish brown to brownish orange. Odors and exudates absent.

Colonies on OMA attaining 25-33 mm diam. in 5 weeks at 22°C. Margin uneven, appressed. Aerial mycelium crustose to felty, irregularly wrinkled, but not as much as on PDA. Colony not raised at the center, greyish orange or brownish orange. Odors and exudates absent.

Colonies on CMA attaining 19-23 mm diam. in 5 weeks at 22°C. Margin even, submerged. Aerial mycelium almost absent, although the presence of sporulation areas confers the colony a finely granular appearance. Colony mainly colorless, except for the sporulating areas close to the center, which appear greyish orange to brownish orange or light brown. Reverse greyish orange to brownish orange or light brown at the center, translucent towards the margins. Odors and exudates absent.

Discussion

According to the last revision of the genus *Titaea* (Sutton, 1984), the name *Titaea* would be restricted to four species of hyperparasitic hyphomycetes with hyaline and morphologically complex conidia: *T. callispora* Sacc. (type species), *T. clarkeae* Ell. & Ev., *T. doidgeae* Hansf. and *T. hemileiae* Hansf. Recently, Matsushima & Matsushima (1996) have described two new species, *T. volucriata* K. Matsushima & T. Matsushima, and *T. complexa* K. Matsushima & T. Matsushima, both isolated from decaying plant material.

Titaea formosa sp. nov. superficially resembles two of the four species accepted by Sutton (1984), namely *T. hemileiae* and *T. doidgeae*. The three species have bilaterally symmetrical conidia consisting of six elements, in which two upper lateral branches are clearly longer than the rest. However, the size and shape of the cells differ significantly amongst the species.

The elongated elements in the conidia of *T. doidgeae* are 1-2 septate, filiform and very long (32-46 µm) compared with the other two species. With respect to *T. hemileiae*, there is a clear

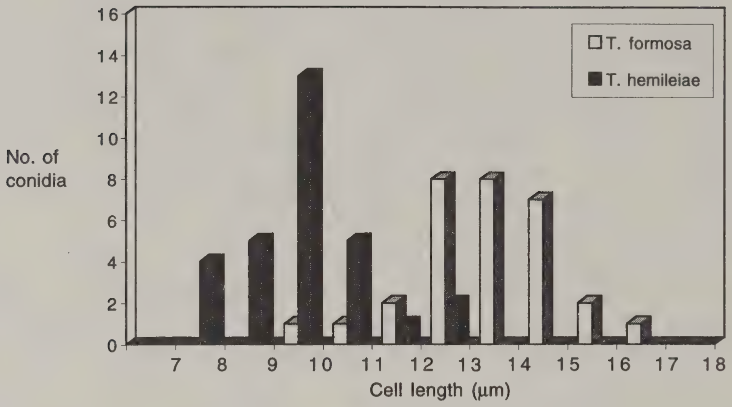
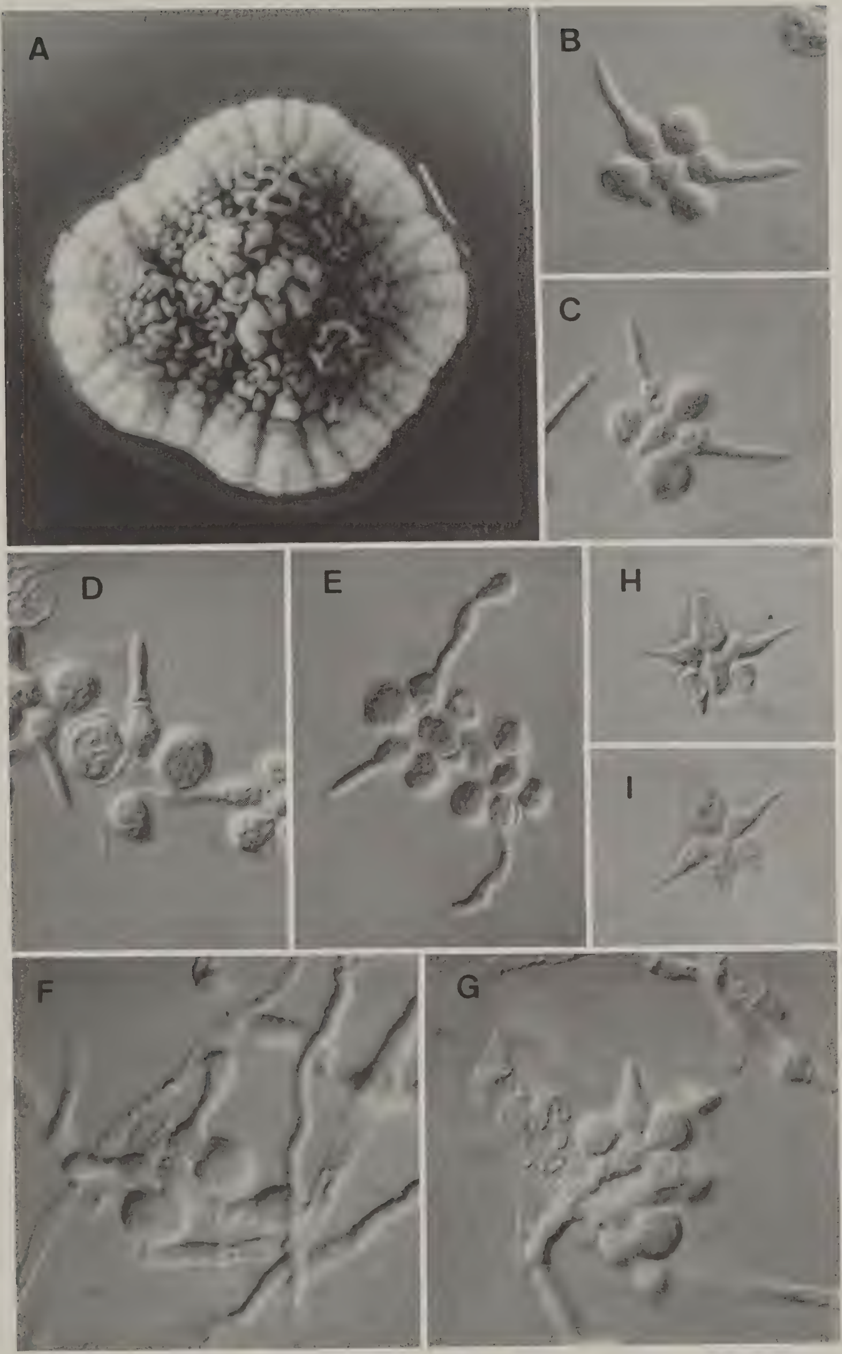


Fig. 3. Length distribution of the upper lateral conidial elements, based on 30 randomly selected conidia of *Titaea hemileiae* (IMI 5007, holotype) and *Titaea formosa* (MA-37927, holotype).

difference in shape of the apical and the two lower lateral cells, which are conical-ovoid in *T. hemileiae* (figs 2H, 2I), and globose in *T. formosa* (figs 2B, 2C). Moreover, the size of the lageniform upper lateral elements is different in both species. The original description of *T. hemileiae* by Hansford (1946) gives a range in length of 10-12 µm (7-11 µm according to Sutton, 1984). We have measured 30 conidia randomly selected from Hansford's original herbarium material, determining that the length distribution follows a curve centered in the 9-10 µm interval, with a total range of 7-13 µm (figure 3). When the same number of conidia of *T. formosa* were measured, the distribution was centered in the 12-14 µm interval, spanning from 9 to 17 µm (fig. 3). It can be seen that the overlap between both distributions is limited. Lastly, in *T. hemileiae* the longer elements remain always aseptate, whereas in *T. formosa* they may occasionally become septate near the base (figs 2D, 2E). All these differences are relevant enough to consider *T. formosa* as a different species from *T. hemileiae*.

Figure 2. A-G: *Titaea formosa*. A. Five-week-old culture on PDA (x3). Note the cerebriform appearance of the colony center. B, C. Conidia (x1250). D, E. Overmature conidia (x1250). Note the septum near the base of the lageniform lateral element and the germinating cells. F, G. Conidiogenous cells with attached conidia (x1250). H, I. Conidia of *Titaea hemileiae* IMI 49981 (b) (x1250). Note the shape of the apical and lower lateral cells and the conidial size.


A
B
C
D
E
H
I
F
G

The key to the accepted species of the genus *Titaea* proposed by Sutton (1984) can be modified as follows to include the present species and the ones described by Matsushima & Matsushima (1996):

1. Conidia consisting of more than six elements...........***T. complexa***
1. Conidia consisting of six elements or less..2
2. Conidia bilaterally symmetrical, consisting of six elements...3
2. Conidia asymmetrical or symmetrical, consisting of less than six elements...5
3. Upper lateral elements filiform, >30 µm long, septate and divergent...***T. doidgeae***
3. Upper lateral elements long conic or lageniform, < 20 µm long, usually aseptate..4
4. Apical and lower lateral cells conic, upper lateral elements mostly 7-11 µm..***T. hemileiae***
4. Apical and lower lateral cells globose, upper lateral elements mostly 12-15 µm...***T. formosa***
5. One lower lateral cell subulate, downwardly directed, the other not developed..***T. callispora***
5. Lower lateral cells absent...6
6. Upper lateral elements septate, filiform, > 30 µm long, widely divergent...***T. clarkeae***
6. Upper lateral elements aseptate, < 25 µm long, often strongly curved..***T. volucriata***

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MYCOTAXON

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A NEW SPECIES OF PLEUROTHECIOPSIS FROM A RAINFOREST IN VENEZUELA

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ABSTRACT

Pleurotheciopsis sylvestris anam. sp. nov., collected on rotten leaves of an unidentified plant in a Venezuelan rainforest, is described and illustrated. This species is characterized by navicular to broadly fusiform, 4–7 (mostly 5)–septate conidia with pale brown to yellow central cells and hyaline to subhyaline end cells. Notes on closely related species are given.

Key words: *Pleurotheciopsis,* Hyphomycetes, leaf litter, systematics, tropical fungi, Venezuela.

TAXONOMIC PART

Pleurotheciopsis sylvestris R. F. Castañeda et. Iturriaga anam. sp. nov.
Figs. 1–6.

Etymol. L. *sylvestris*–refers to its growing among trees, pertaining to woods.

Ad fungus conidiales, hyphomycetes pertinens. ***Coloniae*** *in substrato naturali pilosae, effusae, hypophyllae, atrobrunnea usque ad brunnea. Mycelium partim superficiale et partim in substrato immersum, ex hyphis septatis, ramosis, laevibus, brunneis, 2.5–4.0 µm diam. compositum.* ***Conidiophora*** *macronematosa, mononematosa, erecta, recta, 10–15–*

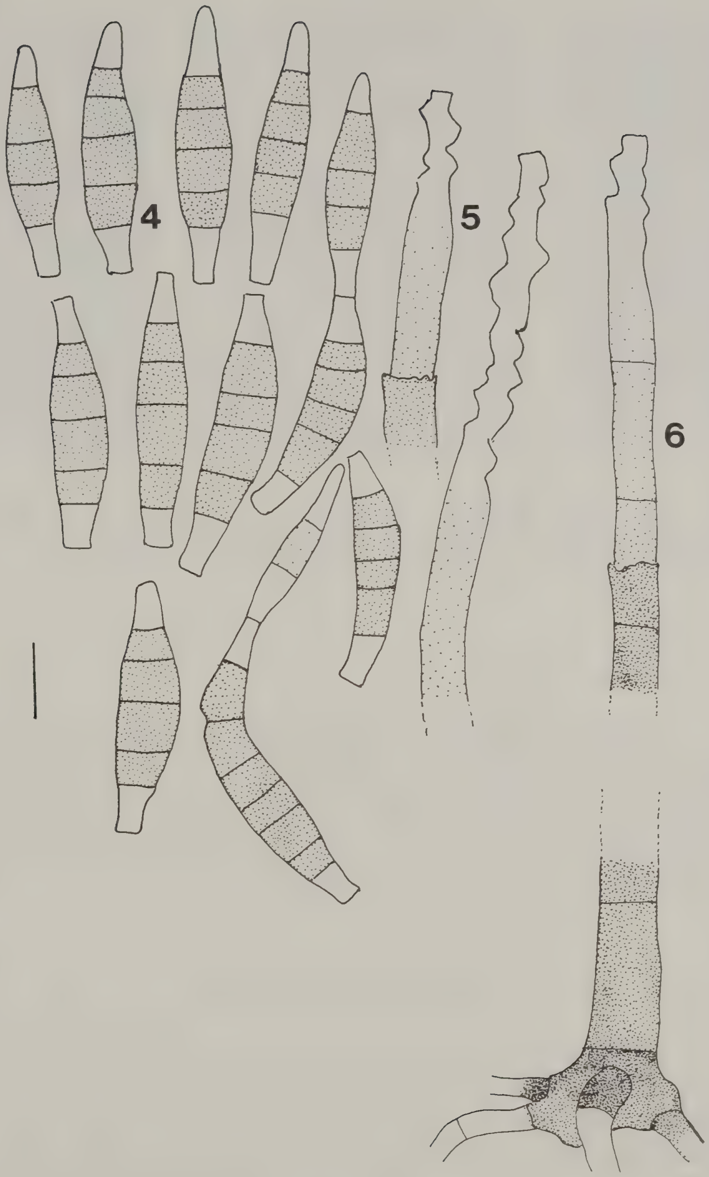
septata, non ramosa, 200–350 x 12–18 µm, atrobrunnea ad basim, subhyalina vel hyalina ad apicem, interdum cum 1–2 proliferationibus percurrentibus. ***Cellulae conidiogenae*** *terminales, sympodiales, polyblasticae, denticulatae, cum cicatricibus declivibus; subhyalinae usque ad hyalinae, 30–50 x 7.0–7.5 µm, in conidiophoris incorporatae.* ***Conidiorum secessio*** *schizolytica.* ***Conidia*** *holoblasticae, blastocatenata navicularia usque ad lata fusiformia, interdum curvata, conidia intercalaria utrimque truncata, 4–7 (plerumque 5)–septata, 35–49 x 8.0–8.5 µm, conidia apicalia truncata ad basim, obtusa ad apicem, 29–41 x 7–8 µm; conidia in cellulis intermediis dilute brunneis usque luteis et utrimque hyalinis.Teleomorphosis ignota.*

Substratum: *In foliis dejectis non determinatae.*
Habitat: *In sylva pluvialis.*

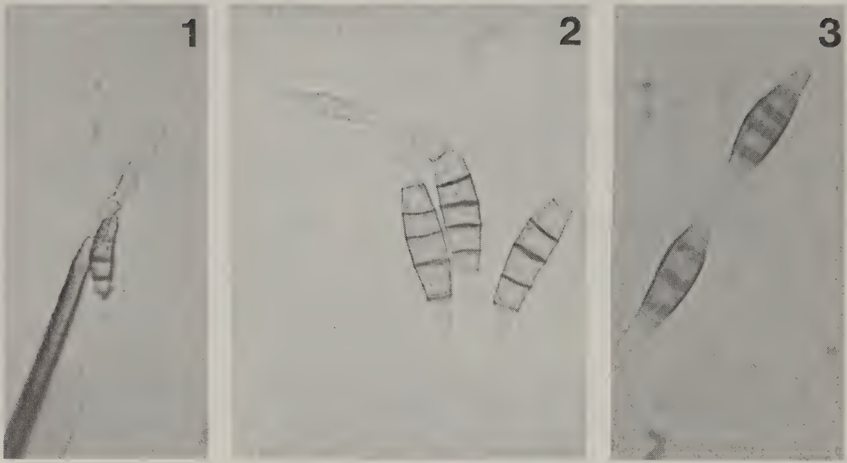
HOLOTYPE: VENEZUELA, ESTADO ARAGUA: Camino de Interpretación de la Naturaleza "Andy Fields", Estación Biológica Rancho Grande, Parque Nacional Henry Pittier, 1200 m altitude. Leg. R. F. Castañeda & T. Iturriaga, 25.XI.1997, (MUCL 40695).

Conidial fungi, Hyphomycetes. **Colonies** on natural substrate hairy, effuse, hypophyllous, dark brown to brown. Mycelium partly superficial and partly immersed. Hyphae septate, branched, smooth–walled, brown, 2.5–4.0 µm diam. **Conidiophores** macronematous, mononematous, erect, straight, 10–15–septate, unbranched, 200–350 x 12–18 µm, dark brown at the base, subhyaline toward the apex, sometimes with 1–2 percurrent proliferations. **Conidiogenous cells** terminal, integrated sympodial, polyblastic, irregular with broad, flat, sloping scars, subhyaline to hyaline, 30–50 x 7.0–7.5 µm. **Conidial secession** schizolytic. **Conidia** holoblastic, blastocatenate, navicular to fusiform, sometimes curved, intercalary conidia truncate, 4–7–(mostly 5)–septate, 35–49 x 8.0–8.5 µm, apical conidia truncate at the base, obtuse at the apex, 29–41 x 7–8 µm; intermediate cells pale brown to yellow, end cells hyaline to subhyaline. Teleomorph unknown.

Substrate: On fallen, decaying leaves of an unidentified plant.
Habitat: Rainforest at Rancho Grande Biological Station, Andy Fields Nature Interpretation Path, Henry Pittier National Park, 1200 m altitude, Aragua State, Venezuela.



FIGS. 1–3. ***Pleurotheciopsis sylvestris.*** 1. Conidiogenous cell and conidium. 2. Conidia. 3. Two attached conidia. 1 x400, 2 & 3 x1000.



FIGS. 4–6. ***Pleurotheciopsis sylvestris.*** 4. Several detached conidia, and two pairs still connected. 5. Sympodial conidiogenous cells. 6. Conidiophore showing percurrent proliferation. Scale bar = 10 μm.

The three previously described species of ***Pleurotheciopsis*** B. Sutton have hyaline conidia: ***P. pusilla*** B. Sutton (1973) (type species of the genus) has 1–2–septate conidia, 6.5–12.5 x 2.0–2.5 µm ; ***P. bramleyi*** B. Sutton has 2–3–septate conidia, 16.5–25 x 4.5–6.5 µm. ***P. setiformis*** R. F. Castañeda (1985) has both conidiogenous cells and setae, and might represent a distinct undescribed new genus.

ACKNOWLEDGMENTS

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FOMITOPSIS CAJANDERI IN EUROPE - EVIDENCE FROM MATING EXPERIMENTS.

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*Corresponding author

Abstract

The occurrence of *Fomitopsis cajanderi* (Karst.) Kotl et Pouz in Europe is confirmed by mating experiments. In this study we report the interfertility among European and North American isolates of *F. cajanderi* and the intersterility among sympatric European *F. cajanderi* and *Fomitopsis rosea* (Alb. et Schw. Fr.) Karst. isolates. The taxonomical and ecological implications of the results are discussed.

Key words: Basidiomycotina, Polyporaceae, *Fomitopsis cajanderi*, Mating test

Interfertility tests is a useful criterion to separate morphologically similar species (Ryvarden & Gilbertsson 1993). *Fomitopsis cajanderi* (Karst.) Kotl et Pouz and *Fomitopsis rosea* (Alb. et Schw. Fr.) Karst. are two species with similar macro- and micromorphology separated by complete absence of interfertility between homocaryons (Mounce & MacRae 1937).
Fomitopsis cajanderi is perennial with a sessile effused-reflexed basidiocarp (Ryvarden & Gilbertsson 1993). The upper surface is light brown to pinkish in young specimens darkening to pinkish brown or gray to blackish in old specimens (Ryvarden & Gilbertsson 1993) and it produces a brown cubical rot in dead conifers. The sexual system of *F. cajanderi* is heterothallic and bipolar (Mounce and MacRae 1937). It is widely distributed in the forest regions of North America, Japan, eastern Siberia and Mongolia but rare in Europe (Ryvarden & Gilbertsson 1993). *F. rosea h*as more or less the same morphological characters as *F. cajanderi* (Ryvarden & Gilbertsson 1993). It has a circumboreal distribution and in Europe and it decayes Picea and rarely *Populus*, *Alnus* and *Pinus* with a brownrot.
Both species belong to the *Fomitopsis rosea* species complex where the species can be separated by mating tests or by careful cultural studies of growth rate at optimum temperatures (Carranza-Morse & Gilbertsson 1989). The latter authors also reported interfertility among two isolates of *F. cajanderi* from Mexico and Idaho respectively. Moreover, *Fomitopsis feii* (Fr.) Kreisel, another species in the *Fomitopsis rosea* complex was found to be intersterile among Asian and American isolates.
In this study we tested the interfertility between European and North American isolates of *F. cajanderi* and the interfertility between European isolates of *F. cajanderi* and *F. rosea.*

Material and Methods

The isolates used in this study are listed in table 1. *F. cajanderi* was sampled in the botanical reserve, Syktyvkar, Republik of Komi, Russia and The Sierra Nevada Mountains, California, United States of America and *F. rosea* the Kotin Forest in Lammi, Finland and in the botanical reserve, Syktyvkar, Republik of Komi, Russia. Basidiospores were collected on glass plates that were placed under the sporocarp either directly in the field or in the laboratory with a wet piece of cloth around. To avoid resampling of the same genet , all sporocarps were from separate logs. The basidiospores were suspended in water and spread onto modified Hagem agar plates (Högberg et al 1995). Between 6-15 germinated spores were individually transferred, with a dissecting microscope, to new media to generate haploid single spore cultures. All isolates were morphologically carefully checked that clamp connections were absent to verify that homokaryons were used for the mating tests.

Table 1. List of isolates used in the interfertility tests.
All isolates are now deposited at the Department of Forest Mycology & Pathology, Swedish University of Agricultural Sciences Uppsala.

Isolate	Species	Origin
Sy 1-c	*F. cajanderi*	Syktyvkar, Republic of Komi, Russia
Sy 1-d	*F. cajanderi*	Syktyvkar, Republic of Komi, Russia
Sy2-a	*F. cajanderi*	Syktyvkar, Republic of Komi, Russia
Sy2-g	*F. cajanderi*	Syktyvkar, Republic of Komi, Russia
SN2-8	*F. cajanderi*	Sierra Nevada, California, USA
SN2-7	*F. cajanderi*	Sierra Nevada, California, USA
SN1-2	*F. cajanderi*	Sierra Nevada, California, USA
SN1-7	*F. cajanderi*	Sierra Nevada, California, USA
Frk1-b	*F. rosea*	Lammi, Finland
Cr16b-d	*F. rosea*	Syktyvkar, Republic of Komi, Russia
Cr1d-d	*F. rosea*	Syktyvkar, Republic of Komi, Russia

The mating types from each sporocarp of *F. cajanderi* were determined by mating five homocaryons from each sporocarp in all possible combinations (data not shown). One mating type each of the homocaryons of *F.rosea* were used as tester strains for interfertility with *F. cajanderi*. The mating types of the isolates representing *F. rosea* were determined in an earlier study (Högberg et al 1997). Isolates tested for interfertility were placed on agarplates at an approximate distance of about four centimetre and left for growth and heterokaryotisation for five weeks. Then, the presence of clamp connections were checked under microscope. In a second step, two isolates from each sporocarp of *F. cajanderi*, representing different mating type genes, and three homokaryon tester strains of *F. rosea* were crossed in all possible combinations.

Result and Discussion

All isolations of homokaryons were successful as none of the isolates had clamp connections. The mating types within the four sporocarps of *F. cajanderi* segregated as expected. The results of the interspecific interfertility tests are shown in Figure 1.

None of the *F. cajanderi* isolates are crossfertile with *F. rosea* isolates. This is in agreement with earlier results (Mounce & MacRae 1937). All intraspecific parings of *F. cajanderi* were compatible except the pairing between SN2-8 and SN1-7 that did not show clamp connections in the culture. This exception could be due to repeated sampling of the same mating type in a local area or be an error during the microscopical examination. The complete fertility between European and American isolates of *F. cajanderi* is firm evidence for the presence of *F. cajanderi* in Europe that confirm earlier reports from Europe and Morocco of this species. The ecology of *F. cajanderi* may indicate why this fungus is rare in Europe compared to *F. rosea*. In the Republikc of Komi in Russia *F. cajanderi* is found only on logs that are flooded during long periods of the late spring, an environment where *F. rosea* is not found. This may indicate competitive exclusion of *F. cajanderi* in Komi forests not subjected to regular floods where *F. rosea* is frequently found on coarse woody debris.

Figure 1. Result of mating tests among sympatric European isolates of *F. cajanderi* and *F. rosea* and allopatric mating tests of *F. cajanderi*.
+ indicate presence and - indicate absence of clamp connections.

	Sy1-c	Sy1-d	Sy2-a	Sy2-g	SN2-8	SN2-7	SN1-2	SN1-7	Frk1-b	Cr16bd	Cr1d-d
Cr1d-d	-	-	-	-	-	-	-	-	+	+	-
Cr16bd	-	-	-	-	-	-	-	-	+	-	
Frk1-b	-	-	-	-	-	-	-	-	-		
SN1-7	+	+	+	+	-	+	+	-			
SN1-2	+	+	+	+	+	+	-				
SN2-7	+	+	+	+	+	-					
SN2-8	+	+	+	+	-						
Sy2-g	+	+	+	-							
Sy2-a	+	+	-								
Sy1-d	+	-									
Sy1-c	-										

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Many thanks to Tom Bruns, UC Berkeley for the sampling and transfer of basidiospores from the two specimen of *F. cajanderii* sampled in the Sierra Nevada mountains, California, USA. We also appreciate the comments from Leif Ryvarden, University of Oslo on an earlier draft of this manuscript. Financial support for this work was received from The Swedish institute, The Oscar and Lili Lamm foundation and the Swedish Royal Academy of Sciences.

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PENICILLIUM DONKII IN ANTARCTIC POND

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ABSTRACT - *Penicillium donkii* Stolk MULC 39880 was isolated from sample of mud collected from Gondwana Lake, Antarctica. This strain, like the type species isolated from soil in Alaska, produces soft, reddish brown, sclerotia.

KEY WORDS - *Penicillium donkii*, Antarctica.

The fungi isolated from samples collected from mud of Antarctic ponds, situated between latitude 72° and 74° S and longitude 162° and 169° E in continental Antarctica were investigated (Montemartini and Gestro, 1994) as part of the National Program of Antarctic Researches (PNRA). These investigations were continued during the campaign 1993/94. Here we report the isolation of *Penicillium donkii* Stolk MULC 39880 from a sample of mud collected from Gondwana Lake, Antarctica by S. Onofri. This small lake is situated at 74° 36' S and 164° 56' E between Gerlache inlet (Terra Nova Bay) and Gondwana Station. This lake, like others described by Guilizzoni et at. (1991), has a low biological activity, elevate pH and scarce CO_2 due to the presence of a thick ice cover, which lasts for the majority of the year. The salt (NaCl) content is high, for the result of wind driven sea spray, total phosphorus is elevated, the nitrogen is scarce.

Our strain of *P. donkii* was compared with the type strain (MUCL 31188; Fig. 1), isolated from arable soil in Alaska (Stolk, 1973), and the type strain of *P. thomii* Maire (MUCL 31204), using the diagnostic criteria of Pitt (1979) and Stolk (1973). Like the type strain of *P. donkii*, the Antarctic strain is monoverticillate (Figs. 2 & 3), produces small subglobosa conidia (2 - 2,3 - 3 x 2 - 2,2 - 2,4 µm) and soft, reddish-brown sclerotia. Sclerotia produced by the Antarctic strain were larger (up to 200 µm in diameter), than those produced by the type strain of *P. donkii*. These sclerotia are readily distinguished from the hard, pink-colored sclerotia produced by *P. thomii* (Raper and Thom, 1949).

The Antarctic strain has more brilliantly pigmented conidia (Methuen 25/26, D 3/4) than the type strain (Methuen 26, C-D 2/3) while the Antarctic has a less brightly pigmented colony reverse ((Methuen 6, C-D 5) when grown on both Czapek yeast autolysate agar and in Czapek agar. We also confirm the ecological preference of *P. donkii* for cold environments, both because strains were isolated in cold habitats and because they grown at low temperature.

Both strains of *P. donkii* were tested for their ability to inhibit the growth of following bacterial strains: *Escherichia coli* ATCC[2] 25922, *Staphylococcus aureus* ATCC 29212, *Micrococcus luteus*_ ATCC 4698, according to Lorian (1991) with negative results.

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[1] This research is supported by National Program of Antarctic Research (PNRA)

[2] ATCC = American Type Culture Collection Rockville, Maryland (20852 U.S.A.)

The Antarctic strain of *P. donkii* has been deposited with the Mycothèque de l'Université Catholique de Louvain la Neuve (MUCL).

ACKNOWLEDGEMENTS

The authors are indebted to the colleague Onofri S., who collected the samples during the campaign 1993/94 of P.N.R.A.

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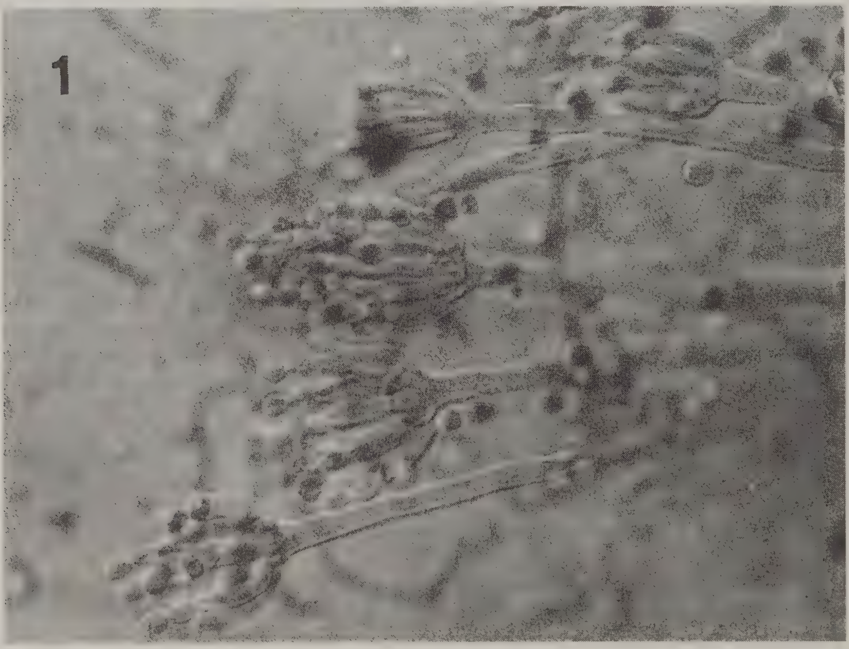
FIGURES

Fig. 1 - Twenty days old colony of type strain (1000X).
Fig. 2 - Twenty days old colony of Antarctic strain (630X).
Fig. 3 - Twenty days old colony of Antarctic strain (1000X).



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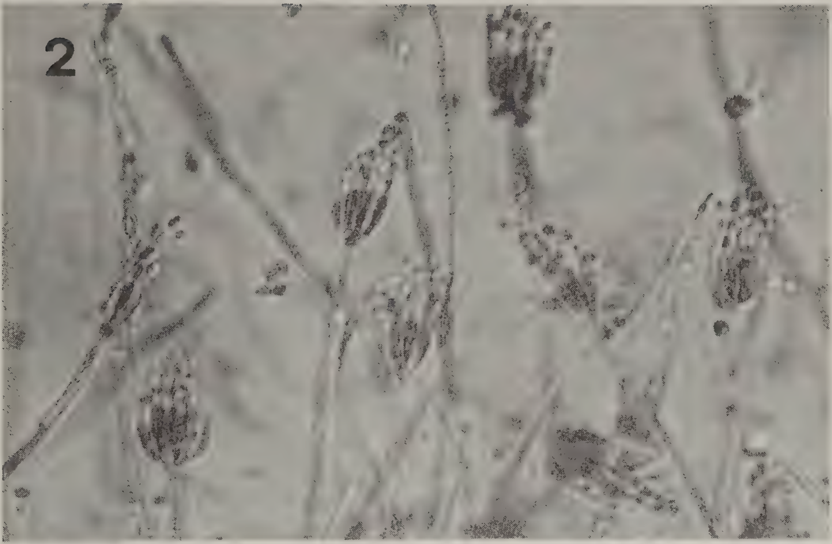

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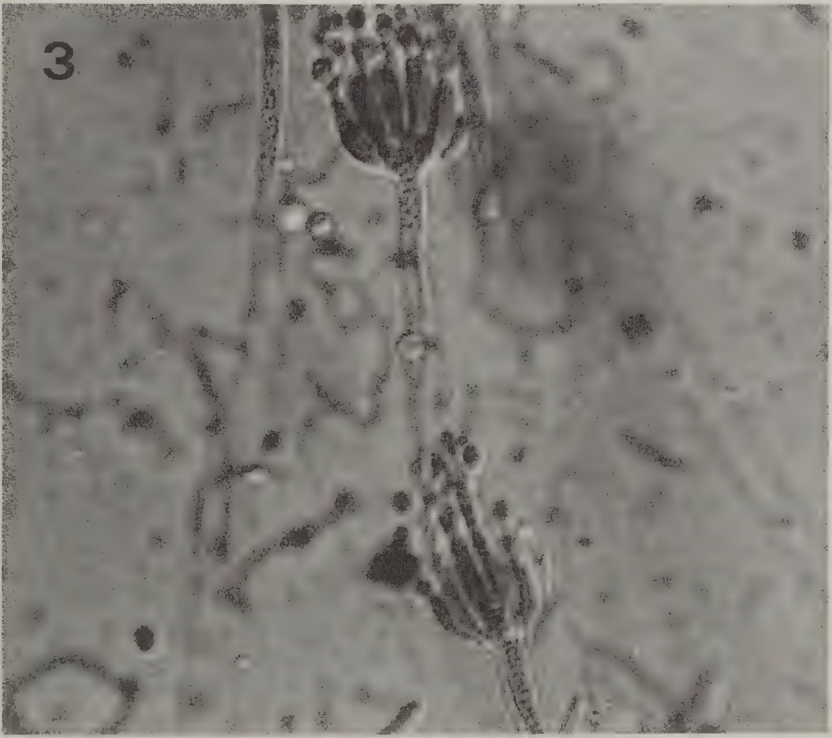

MYCOTAXON

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ADDITIONS TO THE KNOWN XYLARIACEAE (HYMENOASCOMYCETES, XYLARIALES) OF SONORA, MEXICO

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ABSTRACT

Xylaria feejeensis in its strict sense constitutes the first record to México. **Daldinia eschscholzii** and **X. poitei** are reported for the first time from Sonora state. Teleomorphic and anamorphic data of **X. striata**, a taxon previously reported from Argentina (as **X. deserticola**) and China, are given. **Xylaria striata** var. **sonorensis** var. nov. is proposed on the grounds of its smaller stromata and shorter ascospores, as compared with the typical variety.

Key words: **Daldinia**, Mexico, Systematics, **Xylaria**, Xylariaceae.

There is only one paper devoted to Xylariaceae from Sonora state, México. In that work Pérez-Silva et al. (1996), reported 16 taxa from 16 collecting sites. With this contribution the known Xylariaceae of Sonora is augmented by reporting **Xylaria feejeensis** *sensu stricto*, **Daldinia eschscholzii** (Ehrenb.: Fr.) Rehm, **Xylaria poitei** (Lév.) Fr., and **Xylaria striata** var. **sonorensis** var. nov. By using data of authentic material and Mexican collections, a complete description of **Xylaria striata** Pat. is provided.

The field work was done in Los Alamos-Arroyo Cuchujaqui, in the municipality of Los Alamos, state of Sonora. Los Alamos-Arroyo Cuchujaqui is situated in the Sierra Madre Occidental and its coordinates are 27° 01' Lat. N; 108° 55' Long. W. There are in the place the following plant communities: low deciduous tropical forest, oak forest, and oak-pine forest (Rzedowski, 1978). The low deciduous tropical forest is considered the northern boundary of such a plant association in the American Continent, and the dominating plant species are **Bursera confusa** (Rose) Bull.,**B. inopinnata** Bull., **Haematoxylon brasiletto** Karst., **Ceiba acuminata** (Wats.) Rose, **Lysiloma watsoni** Rose, **Taxodium mucronatum** Ten., **Ficus** spp., **Guazuma ulmifolia** Lam., and **Vitex mollis** H. B. K. (Jenkins et al., unpublished data).

The modes of collecting, preservation, and identification of taxa follow San Martín and Rogers (1995). Cultures and anamorphic information were obtained as given in San Martín and Rogers (1993). Herbarium acronyms are largely based on Holmgren et al. (1990).

TAXONOMIC PART

Daldinia eschscholzii (Ehrenb.: Fr.) Rehm, Ann. Mycol. 2: 175. 1904.

The material is as described by Ju et al. (1997).

SPECIMENS EXAMINED: 14.09.1997, Esqueda 3361 and 3392, on dicot wood (CESUES, ITCV); 15.09.1997, Ana Núñez 3522, on wood (CESUES, ITCV); Esqueda 3502, on wood of **Senna atomaria** L. (CESUES, ITCV).

ADDITIONAL MATERIAL EXAMINED: Philippines, Luzon, Laguna Province, Los Baños, Baker, C. F. 516, wood (BPI 716999 [Lloyd Herb. 12401] LECTOTYPE).

NOTES: For lectotypification of **D. eschscholzii** see Ju et al. (1997). The Sonoran material is typical of the species.

Xylaria feejeensis (Berk.) Fr., Nova Acta Regiae Soc. Sci. Upsal. (Ser. 3) p. 128. 1851.

Fig 1. A-D

Stromata unbranched to branched, cylindrical to cylindric-clavate with acute to obtuse, sterile or fertile apices, on short to long tomentose stipes that arise from pannose bases, 1.5 cm total length x 1-3 mm diam; externally blackish with brown outer layer split in polygonal plaques, internally white. Texture woody. Surface rugose and roughened with ostiolar papillae. **Perithecia** 0.2-0.3 mm diam. Ostioles papillate, conspicuous and crowded. **Asci** eight-spored, cylindrical, long-stipitate, 90-125 µm total length x 5-6 µm broad, the spore-bearing part 60-65 µm, with apical ring bluing in Melzer´s iodine reagent, somewhat discoid, 0.75 µm high x 1.5 µm broad. **Ascospores** brown to dark-brown, ellipsoid with narrow ends, with one end sometimes bearing a tiny cellular appendage, 8-9 (-10) x 4.5-5 µm, with straight germ slit spore-length.

Colonies covering Petri dish in 3 wk, at first white with dark-green shades, with entire margins, zonate, floccose, later producing alternately white and dark-green rings from which superficial, black and tomentose, hump-like stromatal primordia arise. Reverse brownish. Conidium-bearing regions on upper surfaces of very reduced stromata. **Conidiophores** in upright palisades, branched or sparingly branched from bases, hyaline, smooth. **Conidiogenous cells** terminal, cylindrical, 34-37 x 3-4 µm, bearing terminal and lateral conidial secession scars. Conidia produced holoblastically in sympodial sequence. **Conidia** hyaline (gray to greenish in mass), smooth, ellipsoid, 10-12 x 4-4.5 (-5) µm, with flattened bases indicating former points of attachment to conidiogenous cells. Some conidia bearing corona-like processes on the apex. C2onidia germinating in water by producing a polar germ tube.

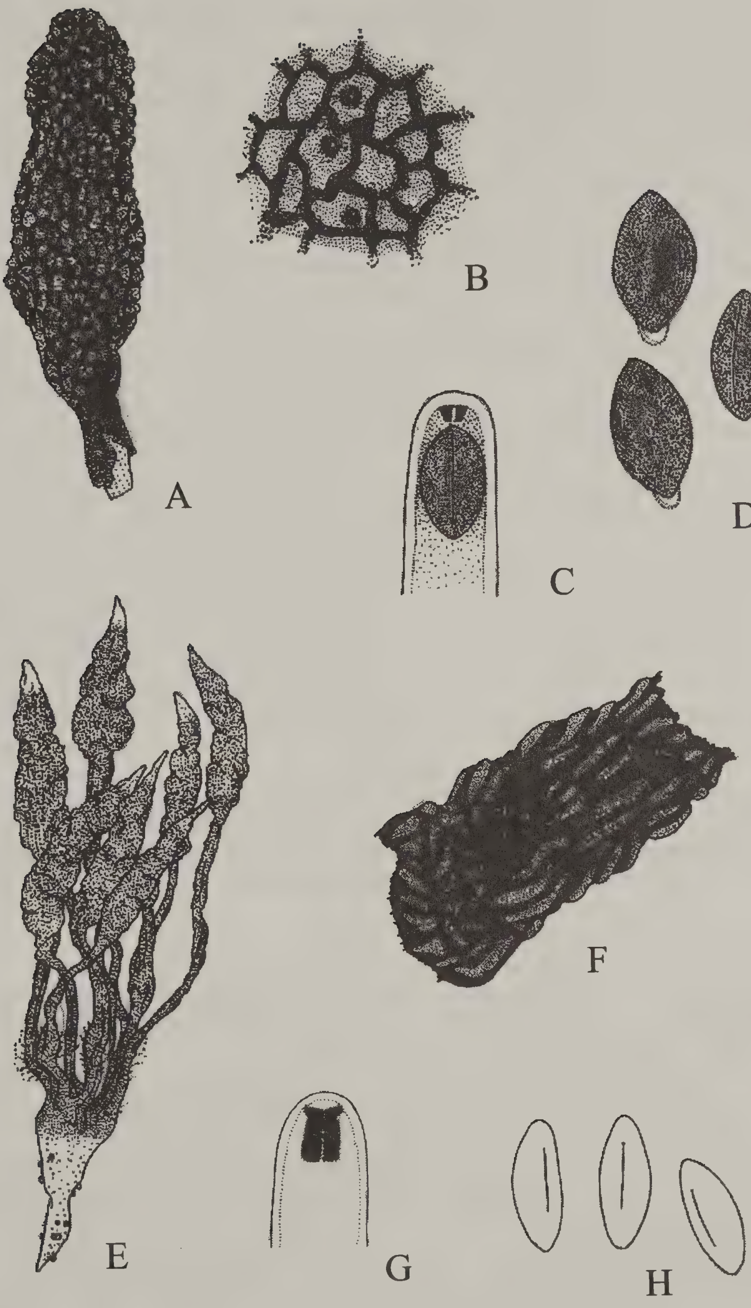
SPECIMEN EXAMINED: 15.09.1997, Esqueda 3500, on dicot wood (CESUES, ITCV).

ADDITIONAL SPECIMENS EXAMINED: Fiji Islands, Hinds, R., no date, (K, HOLOTYPE). México, Veracruz state, Catemaco municipality, Los Tuxtlas Universidad Nacional Autónoma de México Biological Station, 350 m, 4.XI.1988, San Martín 1417 (CULTURED), on wood in evergreen tropical forest (ITCV, JDR).

NOTES: Data of the description agree with the **X. feejeensis**´ protologue (and see Saccardo, 1882) and its description given by Dennis (1956).

Xylaria poitei (Lév.) Fr., Nova Acta Regiae Soc. Sci. Upsal. (Ser. 3) 1, p. 125. 1851.

Fig. 1. A-D: **Xylaria feejeensis** *sensu stricto*. A. Stroma, 1 cm = 3 mm. B. Stroma detail, 1 cm = 0.5 mm. C. Ascus apical ring, 1 mm = 0.35 µm. D. Ascospores, 1 mm = 0.4 µm. E-H: **Xylaria striata** var. **sonorensis**. E. Stromata, 1 cm = 3 mm. F. Stroma detail, 1 cm = 0.4 mm. G. Ascus apical ring, 1 mm = 0.3 µm. H. Ascospores, 1 mm = 0.65 µm.


A
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C
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E
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G
H

The material found in Sonora is as described by Rogers and Callan (1986a) and San Martín and Rogers (1989); the anamorph was described by Callan and Rogers (1986a).
SPECIMEN EXAMINED: 15.09.1997, Esqueda 3484, on dicot wood (CESUES, ITCV).
NOTES: **Xylaria poitei** is distributed in México from semiarid areas in the states of Sonora and Nuevo León to tropical forests of Oaxaca, Veracruz, and Quintana Roo states (San Martín and Rogers, 1989; San Martín, 1992).

Xylaria polymorpha (Pers.: Fr.) Grev., Flora Edin., p. 355. 1824.

Teleomorphic features of the specimens collected in Sonora are as described by Rogers and Callan (1986b) and San Martín and Rogers (1989).
SPECIMENS EXAMINED: 14.09.1997, Esqueda 3361 and 3392, on wood (CESUES, ITCV); 15.09.1997, Esqueda 3460 and 3502, on wood (CESUES, ITCV).

Xylaria striata Pat. var. **sonorensis** San Martín, Esqueda & Pérez-Silva, var. nov.

Fig. 1. E-H

Typical variety in Journ. Bot. 1887. P. 247. 1887.

A varietate typica differt in stromatibus minoribus, i. e., 2-3 cm altus vs 5.5.-9 cm altus et in ascosporis brevioribus, i. e., (11.5-) 12-14 (-15) µm vs 16-18 (-19) µm.

Stroma branched from a long stipes buried deep in the soil, terete to flattened with acute sterile apices, 2.3 cm high x 1.5-1.8 mm broad; externally dark-brown with white or brown outer layer split in bands, internally white, fleshy. Texture rather hard. Surface rugulose with outer layer, ostiolar papillae, and perithecial protrusions. **Perithecia** 0.5-0.6 mm diam. Ostioles discrete to hemispherically papillate. **Asci** cylindrical, eight-spored, with uniseriate to partly biseriate spore arrangement, stipitate to long stipitate, 119-147 µm total length x 5.5-7 µm broad, the spore-bearing part 79-87 µm, with apical ring bluing in Melzer´s iodine reagent, rectangular and more or less inflated at the center, 2.5-3 µm high x 1.5-2 µm broad. **Ascospores** light-brown to brown, transluscent, ellipsoid-inequilateral with narrowly rounded to acute or somewhat pinched ends, (11.5-) 12-14 (-15) x (4.5-) 5 (-5.5) µm, with straight germ slit much less than spore-length on ventral side.
SPECIMEN EXAMINED: 14.09.1997, Esqueda 3439, on wood or roots buried in soil (CESUES; TYPE: ITCV; ISOTYPE).
NOTE: **Xylaria striata** var. **sonorensis** differs from authentic Chinese material in having smaller stromata and shorter ascospores (see next description).

Xylaria striata Pat., Journ. Bot. 1887. p. 247. 1887.
Synonym: **Xylaria deserticola** Speg., An. Mus. Nac. Buenos Aires 8. p. 70. 1902.

Stromata cylindrical, with pointed, sterile apices, several arising from common short to long, smooth and dirty stipes, 5.5-9 cm high x 3-4 mm broad. Externally black with remnants of whitish peeling layer split in longitudinal bands, internally white. Surface somewhat wrinkled with fine papillae. Texture rather hard. **Perithecia** 0.4-0.5 mm diam. Ostioles finelly papillate. **Asci** cylindrical, eight-spored, with obliquely uniseriate to partly biseriate spore-arrangement, very long-stipitate, 137-224 µm total length x 5-7 µm broad, the spore-bearing part 77.5-104 µm, with apical ring bluing in Melzer´s iodine reagent rectangular, 3 µm high x 2.5 µm broad. **Ascospores** light-brown, ellipsoid-inequilateral to navicular with narrow ends, 16-18 (-19) x 4.5-5 µm, with straight germ slit much less than spore-length on concave side.

Colonies covering Petri dish in 2-3 wk, at first white, with entire margins, strongly zonate, velutinous, then with appressed, gray and blackish sectors, finally with radiate, yellowish furrows. Reverse unchanged. **Stromata** produced at the periphery of the colony, at first arising as pale-yellow mycelial tufts, then clavate, unbranched, 1.5-2 cm long x 1-1.5 mm broad, initially white turning blackish from bases upwards, tomentose. Conidium-bearing regions on the upper surfaces of stromata. **Conidiophores** not distinct. **Conidiogenous cells** short-cylindrical, produced as lateral growths from external stromatal hyphae, 5.5-9 (-11) x (2-) 3 µm, yellowish, smooth, bearing terminal denticulate conidial secession scars. Conidia produced holoblastically. **Conidia** hyaline (yellowish in mass), smooth, fusiform to ellipsoid, (5-) 5.5-6 (-6.5) x 2.5-3 µm, with flattened bases indicating former points of attachment to conidiogenous cells.

SPECIMENS EXAMINED: China, Mo So Yu, 1887, collector unknown, monocot wood, **Xylaria striata** Pat. (FH: selected here as the EPITYPE). China, Mo So Yu, Lang Kong, 1887, collector unknown, wood (FH: "TYPE").

ADDITIONAL SPECIMENS EXAMINED: México, Jalisco state, Guadalajara City, 25.IX.1980, Godínez-Anguiano 777, on dead log of **Eucalyptus** sp. (IBUG; ITCV; XAL). Tamaulipas state, Victoria municipality, La Libertad Canyon, XI.1987, Pablo Lavín 24, on wood of a riparian element (ITCV, JDR); Ocampo municipality, El Tigre Hill, 10.VIII.1988, Chacón-Jiménez 429 (CULTURED), on dicot wood in median subdeciduous tropical forest (ITCV; JDR).

NOTES: Type material of **X. striata** at FH is immature. Another package containing specimens from the type locality (Mo So Yu, 1887) has mature authentic material and accordingly, it is selected as the epitype. The Argentinian **X. deserticola** as described by Spegazzini (in Saccardo, 1905) and Dennis (1957), has stromata more robust than those of material described as **X. striata**, but the rest of its characters fit the latter species. Consequently, it is treated here as synonym of **X. striata**. It is noteworthy that in every collection of **X. striata** the stipe is deeply buried in the soil, thus suggesting that the teleomorph comes from immersed plant material.

AKNOWLEDGEMENTS

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XYLARIA PEREZSILVAE SP. NOV. FROM SOIL IN MIXCUM, CHIAPAS. MEXICO

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ABSTRACT

The new species of **Xylaria** (Hymenoascomycetes, Xylariaceae) **X. perezsilvae** is proposed. The taxon was collected from soil in a coffee plantation of ejido Mixcum, Cacahoatán municipality, state of Chiapas, México. Macroscopically the species is peculiar because of the tiny subglobose fertile stroma and the relatively long stipes; microscopically it is characterized by ascospores with a cellular appendage on one end and a hyaline sheath.

Key words: Mexico, Systematics, **Xylaria**.

INTRODUCTION

During the Sixth Mexican Mycological Congress foray to the highlands of Tapachula, Chiapas, a very peculiar **Xylaria** was found growing directly from soil in a coffee plantation. Since the taxon does not appear to be identical with any species of the genus known by the authors, it is described as new.

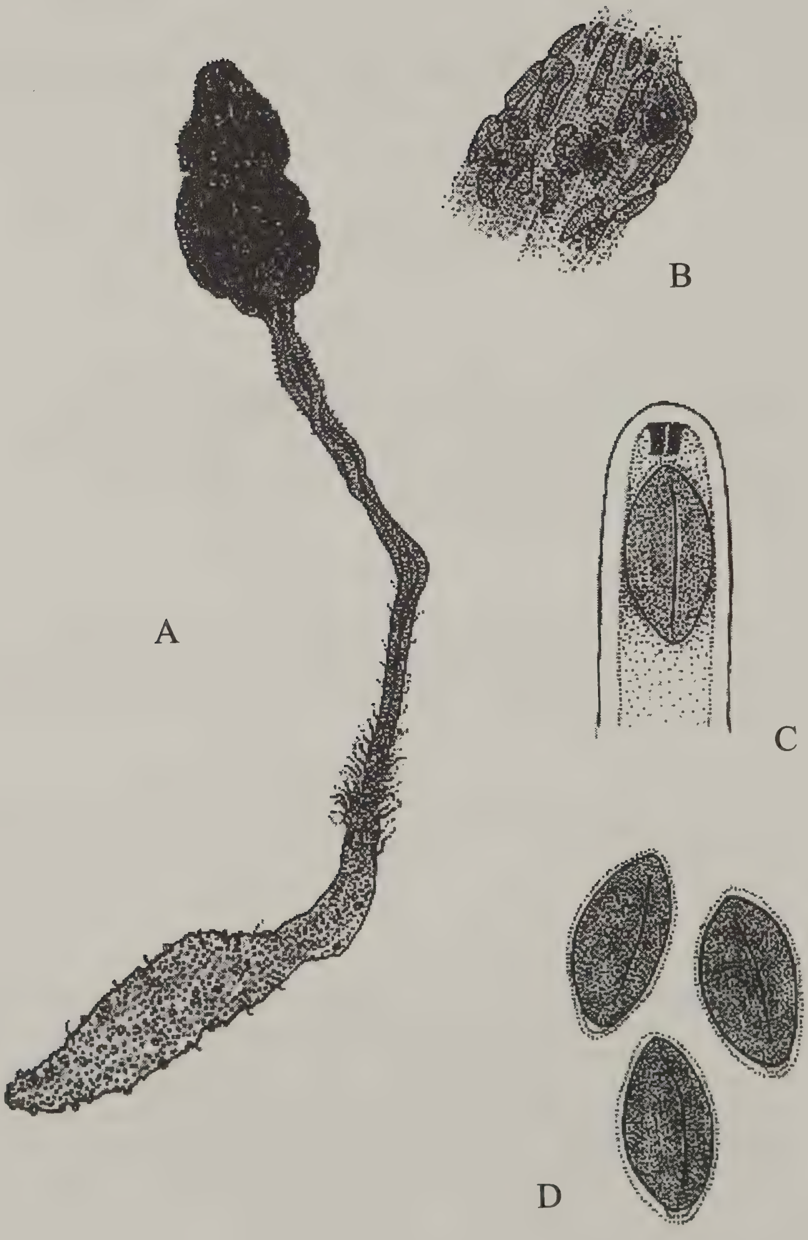
METHODS

The modes of collecting, herborization, and identification of fungal material follow San Martín and Rogers (1995).

RESULTS

Xylaria perezsilvae San Martín & Lavín sp. nov. Fig. 1 A-D

Stromata non ramosa, solitaria, parte fertili subglobosa, tomentosa, 2-2.5 mm altus x 1.3-1.7 mm crassi, protuberationes perithecii plus minusve prominentibus, stipite longo et filiforme, hirsutus, valde sepultus, 2-2.5 cm longo; extus nigra, residuis laminae fuscae facias formantibus, intus alba. Textura plus minusve mollis. Superficies laevis cum tomento, ostiolis et residuis laminae externae. **Perithecia** 0.3-0.4 mm diametro. Ostiola inconspicua vel hemisphaerica. **Asci** cylindrici, octospori, sporis uniseriate, longe sipitati, 93.5-132 µm longitudine tota x 6-6.5 µm crassi, partibus sporiferis 49.5-56 µm, cum annulo apicale in


B
A
C
D

liquore Melzeri cyanescente, quadrato, 2.5 µm altus x 2-2.5 µm crassi. **Ascosporae** brunneae vel fuscae, elipsoideo-inequilaterales cum extremum angustus vel late rotundatis, 8-8.5 (-9) x 4-4.5 µm, cum apendicula hyalina celulare, totae cum vaginis hyalinis indutae et rima germinativa recta per longitudinem sporae, in latus ventralis.

Stromata unbranched, solitary, with subglobose, tomentose fertile part, 2-2.5 mm high x 1.3-1.7 mm broad, with perithecial contours more or less prominent, on long, filiform, hirsute stipes, deeply buried in the soil, 2-2.5 cm long; externally black with brown outer peeling layer split in bands, internally white. Texture more or less soft. Surface smooth, with tomentum, ostioles and outer peeling layer. **Perithecia** 0.3-0.4 mm diam. Ostioles inconspicuous to hemispheric. **Asci** cylindrical, eight-spored with uniseriate spore-arrangement, long-stipitate, 93.5-132 µm total length x 6-6.5 µm wide, the spore-bearing part 49.5-56 µm long , with apical ring bluing in Melzer's iodine reagent, quadrate, 2.5 µm high x 2-2.5 µm broad. **Ascospores** brown to dark brown, ellipsoid inequilateral witnarrowly rounded ends, 8-8.5 (-9) x 4-4.5 µm, bearing a hyaline cellular appendage on one end and a hyaline sheath, with straight germ slit full spore-length.

Etymology. The species is named in honor of Dr. Evangelina Pérez-Silva, the pioneer mycologist in the study of Mexican Xylariaceae.

TYPE: México, Chiapas state, Cacahoatán municipality, ejido Mixcum. Alt. 485 m. 18.X.1997, San Martín 6673Ch, in soil of a coffee plantation.(ITCV, HOLOTYPE).

NOTES: This little **Xylaria** is unlike any species known to the authors. In stromatal morphology it approximates **Xylaria squamulosa** Rogers et San Martín (1989) but the latter grows on wood, has a white outer layer and considerably larger ascospores, i.e., (29-) 31-37 (-40) x (11-) 12-13 (-14) µm vs. 8-8.5 (-9) x 4-4.5 µm. The new species is also reminiscent of **X. mexicana** San Martín et Rogers (1998) but that species grows on leaf veins and petioles, and has larger ascospores, i.e., (9-) 9.5-11.5 (-12) x 5-6 µm vs 8-8.5 (-9) x 4-4.5 µm. **Xylaria perezsilvae** resembles the terricolous **X.** sp. aff. **euphorbiicola** as described by San Martín (1992), but differs in having a subglobose fertile head instead of short cylindrical fertile clavae, and smaller ascospores, i.e., 8-8.5 (9-) x 4-4.5 µm vs 12-13 (-14) x 4.5-5 µm.

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The authors wish recognize the aid of the Sistema Nacional de Investigadores of Mexico, for providing them grants to study the taxonomy and diversity of Mexican Xylariaceae. We thank Dr. Jack D. Rogers, Washington State University, and Dr. John Krug, University of Toronto, for reading the manuscript.

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Fig. 1. **Xylaria perezsilvae**. A. Stroma: 1 cm = 0.5 mm. B. Stroma detail: 1 cm = 0.5 mm. C = Ascus apical ring: 1 mm = 0.5 µm. D. Ascospores: 1 cm = 4 µm.

MYCOTAXON

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MYCENA CALIFORNIENSIS RESURRECTED

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Abstract: *Agaricus* (*Mycena*) *californiensis*, first described by M.J. Berkeley and M.A. Curtis in 1860, represents a common species found in the oak woodlands of California. The species is redescribed, illustrated, and compared with morphologically similar taxa. *Mycena elegantula* Peck is proposed as a synonym.

Key words: Agaricales, California agarics, *Mycena*, taxonomy.

Our current investigation of California species of *Mycena* indicates that *Mycena californiensis* (Berk. & M.A. Curtis) Sacc. is a species common to the coastal oak woodlands of the state. For years, California investigators have collected what was generally identified as *M. elegantula* Peck (1895) or *M. sanguinolenta* (Alb. & Schwein.: Fr.) P. Kumm., on the leaf litter of *Quercus agrifolia* Nee and a few other *Quercus* spp. Upon microscopic examination of these collections, most appeared to share characteristics with *M. elegantula.* One odd character however, was the presence of pigmented laticiferous hyphae throughout the stipe and tramal tissues of all collections. While this character would be expected in those species reported to contain colored latex such as *M. sanguinolenta*, this character has never been reported for *M. elegantula.* Recent collections made by us all contained copious amounts of reddish to orangish red latex.

Investigation of the literature for species superficially resembling *M. elegantula*, revealed the relatively unknown taxon *M. californiensis,* which was described from a single collection made during the U. S. North Pacific Exploring Expedition, 1853-1856. The material was collected by the expedition botanist, Charles Wright, on oak leaves at Mare Island Naval Yard, Solano

Co., California, in January of 1856 (notes with specimen; Pfister, 1978). The collection was presumably sent by M.A. Curtis to M.J. Berkeley in England, and was published as *Agaricus californiensis* Berk. & M.A. Curtis (Berkeley & Curtis, 1860). Unfortunately the published description of the species is very brief, and the existing type specimens are of very poor quality. Smith (1947) included *Mycena californiensis* in the "excluded and doubtful species" portion of his monograph on the genus, stating that the species "cannot be recognized until the microscopic characters of the type are known." Maas Geesteranus (1982, 1992a) investigated the holotype material (K), but was unable to distinguish any defining characters due to the poor state of the collection. Based on his examination of the holotype material, Mass Geesteranus agreed with Smith's view of the species.

We were able to resolve sufficient information from the isotype material of *M. californiensis* [housed at FH(!) and accompanied by Wright's original notes on the material] to conclude that it is conspecific with the commonly encountered oak loving species from California. Investigation of the holotype material of *M. elegantula* revealed that this species is conspecific with the isotype material of *M. californiensis*. Due to the later publication date of *M. elegantula* (Peck, 1895), the binomial *M. californiensis* has priority. The placement of *M. elegantula* as a synonym of *M. californiensis* is therefore proposed.

We present here a more comprehensive description of *Mycena californiensis* based on it's isotype (FH!), on the holotype of *M. elegantula* (NYS!), and on numerous other specimens from California. In the description that follows, color terms and notations are from Kornerup and Wanscher (1978); spore statistics include: $\overline{x}_r$ the range of spore means where $\overline{x}$ is the arithmetic mean of the spore length by spore width for n spores measured in a single sample (specimen), $\overline{x}_m$ the mean of all spores measured (±SD); Q, the quotient of spore length and spore width in any one spore, indicated as a range of variation in n spores measured; $\overline{Q}_r$, the range of $\overline{Q}$-values where $\overline{Q}$ is the mean of Q-values in a single sample; $\overline{Q}_m$, the mean Q-value for all spores measured (±SD). All specimens are deposited at SFSU unless specified otherwise. Frequently cited collectors are abbreviated as follows: D. E. Desjardin (DED), Harry D. Thiers (HDT).

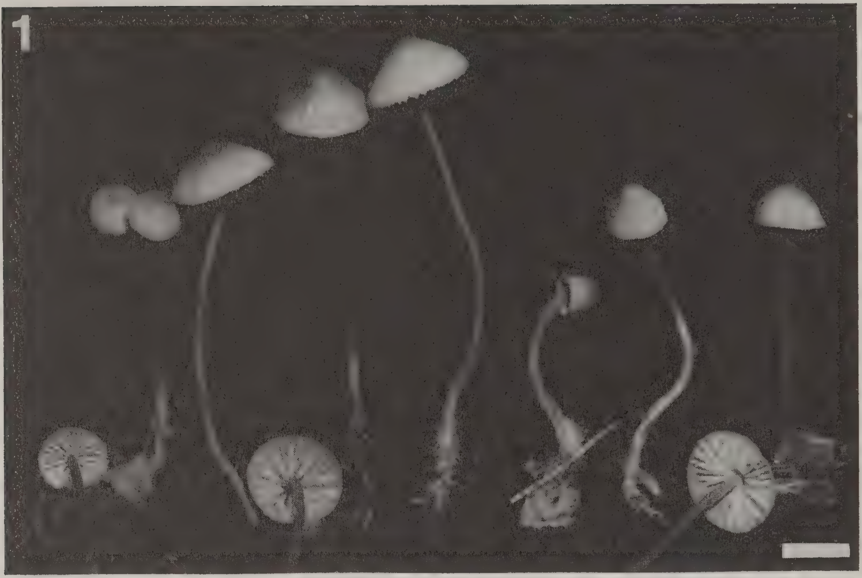
Mycena californiensis (Berk. & M.A. Curtis) Sacc., Syll. Fung. 5: 255. 1887.

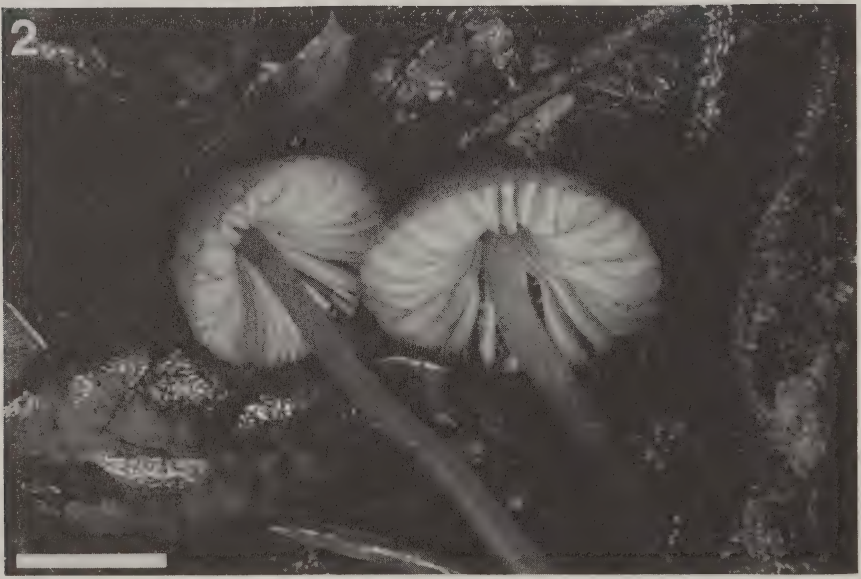
≡*Agaricus californiensis* Berk. & M.A. Curtis, Proc. Amer. Acad. Arts 4: 112. 1860.

= *Mycena elegantula* Peck, Bull. Torrey Bot. Club 22: 199. 1895.

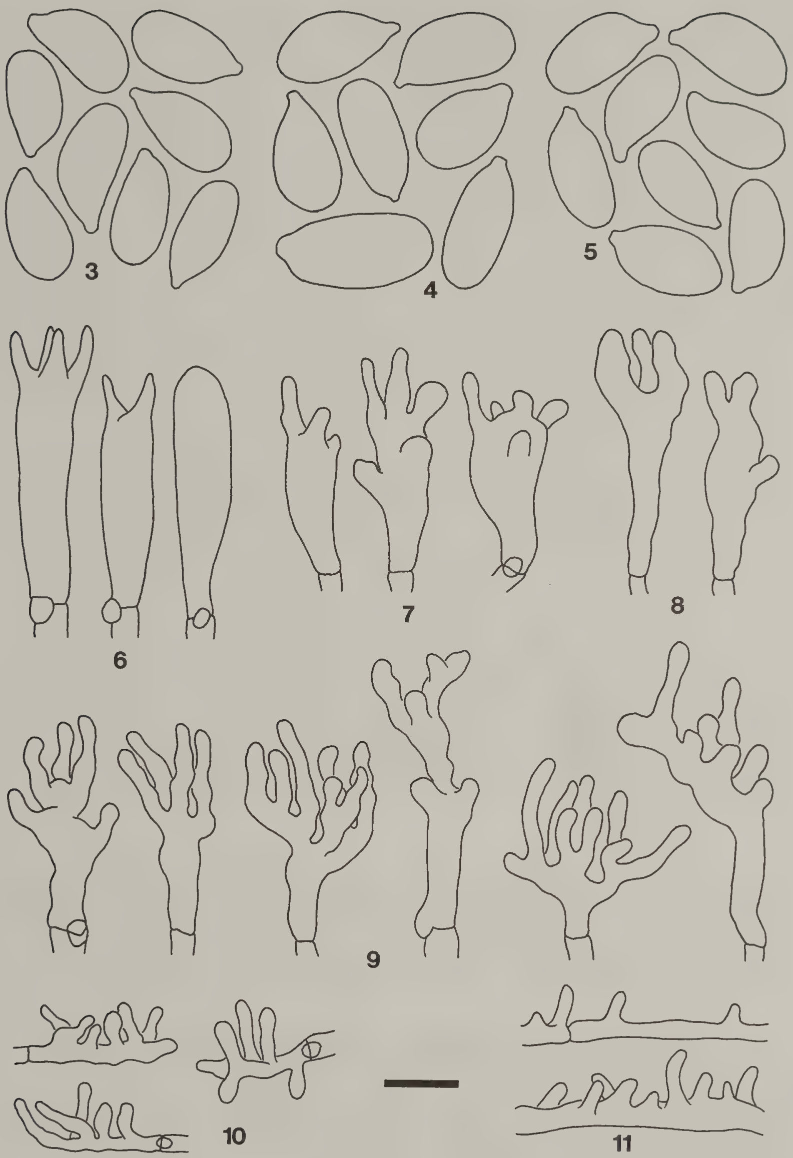
Basidiomes (Figs. 1-2) gregarious to subcaespitose, putrescent. **Pileus** 6-20 mm, obtusely conic to campanulate when young, generally with incurved margins, remaining so with age or expanding to broadly campanulate at maturity, occasionally becoming convex-umbonate; margin entire when young, often becoming wavy or crenate with age, sometimes splitting, striate or striatulate, extreme edges often staining or bruising darkly, pellucid striate; surface dull, moist to dry, glabrous; at first disc dark brown (8F5-6) to reddish brown (9D-F7-8), in age ranging from brownish red (8C6-8) to reddish brown (8D-F5-8), margin generally lighter, grayish red (8C4-6) to reddish brown (8D-E5-7), or brownish orange (7C5-6) when young, in age fading to grayish orange (6B3) or brownish orange (6C3). **Context** thin, concolorous with pileus or paler, often staining dark reddish. **Lamellae** (Fig. 2) ascending adnate to adnate, often with a short decurrent tooth, close to subdistant (15-20) with 1-2 series of lamellulae, moderately broad (1-3 mm), convex; white to pinkish buff, edges reddish brown (9C-D8), reddish orange, or brownish orange. **Stipe** 29-130 X 1-2 mm, central, terete, ±equal, rarely compressed or cleft, dull to shiny, moist to dry, hollow, apex glabrous to pruinose, glabrous below, base covered with strigose to downy tomentum ranging from buff to some shade of orange or pink; apex brownish orange to light brown (7C-D5-8), reddish brown (8D5-6), or grayish red (8C4-6), some fading in age to lighter brownish orange (5-6C4-6), base concolorous or slightly darker; tissue exuding a brownish orange (7C6-8) to reddish brown (9-10D8) or violet brown (11F7-8) latex when cut.

Basidiospores (Fig. 3-5) 8.0-12.0 (-13.0) X 4.0- 6.0 (-6.7) µm [$\bar{x}_r$ = 9.1-11.0 X 4.9-5.6 µm, $\bar{x}_m$ = 9.7 ± 0.8 X 5.2 ± 0.5, Q = 1.4-2.4, $\bar{Q}_r$ = 1.8-2.0, $\bar{Q}_m$ = 1.9 ± 0.2, n = 25 spores per specimen], ellipsoid to amygdaliform, smooth, thin walled, hyaline, weakly to moderately amyloid. **Basidia** (Fig. 6) 26.0-37.5 (- 48.0) X 7.0-10.5 µm, clavate, 4-spored, rarely mixed with 2- spored forms, clamped. **Basidioles** (Fig. 6) clavate,





Figs 1-2. *Mycena californiensis*. 1. Basidiomes (DED 5963). 2. Closeup of lamellar edges (M.Wood s.n.). Scale bar = approximately 10 mm for both figures.



Figs. 3-11. *Mycena californiensis*. 3-5. Basidiospores. 6. Basidia. 7-9. Cheilocystidia. 10. Hyphae of the pileipellis 11. Cortical hyphae of stipe. (Figs. 3,7 - Isotype of *Agaricus californiensis*; Figs. 4,8 - Holotype of *M. elegantula*; Figs. 5,6,9,10,11 - DED 5963) Scale bar = 7.5 µm for Figs. 3-5, 10 µm for Figs. 6-11.

similar to basidia. **Pleurocystidia** absent. **Cheilocystidia** (Fig. 7-9) 16-50 X 6.5-28.0 µm, projections 1.5-18.8 (-30.0) X 1.5-6.5 (-8.0), ranging from contorted-clavate with assorted knob-like projections to irregular in shape, generally with a clavate lower portion and an apical or sublateral portion branching into numerous cylindric projections, these often branching themselves, thin-walled, clamped, with brownish contents that stain darkly in Melzer's reagent. **Pileipellis** (Fig. 10) a cutis; hyphae 1.5-5.0 (-7.0) µm diam, interwoven, covered with abundant cylindric diverticula, 1.5-11.8 (-16.5) X 1.5-2.8 µm, thin-walled, non-gelatinous to subgelatinous, hyaline, inamyloid; terminal cells slightly inflated, otherwise similar to hyphae. **Hypodermium** of inflated cells, 7.0-38.5 µm diam, thin-walled, hyaline, dextrinoid, non-gelatinous; often intermixed with laticiferous hyphae up to 6.5 µm diam, containing brown granular contents. **Hymenophoral trama** dextrinoid, occasionally with laticiferous hyphae as in hypodermium. **Stipe tissues** monomitic; cortical hyphae (Fig. 11) 1.5-6.5 µm diam, parallel, cylindrical, thin walled, covered with scattered to numerous cylindric and occasionally branched diverticula, 2.0-7.0 (-14.0) X 1.0-2.5 µm; terminal cells slightly broader, 3.8-5.0 µm diam, diverticulate; medullary hyphae broader, 4.5-22.0 µm diam, smooth, thin-walled, hyaline; laticiferous hyphae common, 2.8-8.0 (-11.8) µm diam, usually containing brown granular contents. **Clamp connections** observed in all tissues except stipe medullary hyphae.

Habit, habitat and distribution. Gregarious to scattered, often subcaespitose, on leaves and fruits of *Quercus agrifolia*, *Q. lobata* Nee and *Q. kelloggii* Newb. Abundant and common in the coastal oak woodlands of California.

Material examined. California. Contra Costa Co.: Shell Ridge Open Space, 7 Dec. 1996, B.A. Perry 097. Los Angeles Co.: near Pasadena, 22 Dec. 1894, A.J. McClatchie 810 (Holotype of *M. elegantula*: NYS). Marin Co.: Audubon Canyon Ranch, Picher Canyon, 8 Nov. 1979, C. Calhoun 79-910; Audubon Canyon Ranch, Volunteer Canyon, 19 Nov. 1981, C. Calhoun 81-2597; Corte Madera, Deer Run Road, 11 Dec. 1992, DED 5571; Fairfax, Pine Mountain Fire Road, 1 Feb. 1992, DED 5523; Mt. Tamalpais State Park, Lake Lagunitas, 13 Dec. 1993, DED 5963; Olampali State Historic Park, 2 Dec. 1997, B.A.

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of reddish latex, this species is also characterized by fusiform cheilocystidia lacking apical outgrowths. The cheilocystidia of *M. californiensis* are irregular in shape, and have never been observed to be fusiform. Also, all examined California collections of *M. sanguinolenta* appear to be restricted to growth on decaying wood or debris of conifers. *Mycena californiensis* appears to be restricted to the leaves and debris of *Quercus* spp., and has never been observed to be lignicolous. *Mycena purpureofusca* differs from *M. californiensis* in the coloration of the fruiting bodies, which is generally much more violaceous in *M. purpureofusca*, in the absence of pigmented latex, in forming clavate to utriform cheilocystidia, and in growth on coniferous wood. *Mycena renati* has fusiform, subcylindrical and occasionally clavate cheilocystidia, yellow coloration to the stipe, no pigmented latex, and was also reported to grow on conifer wood in the United States (Maas Geesteranus, 1986, 1992b). We have observed that U.S. specimens which are micromorphologically indistinguishable from European *M. renati* lack yellow pigmentation to the stipe and may represent a distinct taxon. *Mycena rubromarginata* (Fr.: Fr.) P. Kumm. is another species that may superficially resemble *M. californiensis*. However, *M. rubromarginata* differs in both habit and cheilocystidia shape, and does not contain colored latex.

It is evident from examination of the type material of *M. elegantula* and the work of Maas Geesteranus (1986, 1992b), that A. H. Smith's concept of *M. elegantula* does not agree with the type material of the species. Smith (1947) reported *M. elegantula* as having clavate to fusoid ventricose cheilocystidia, occasionally with several fingerlike projections, and he reports the species as occurring on decayed conifer wood. Although our investigation of the type material of *M. elegantula* revealed the presence of contorted-clavate cheilocystidia, it contains none that are fusoid-ventricose. The type material of *M. elegantula*, collected from southern California (Los Angeles Co.), is also clearly associated with leaves of *Q. agrifolia*. Investigation of numerous specimens determined by Smith as *M. elegantula*, revealed that his concept of the species (Smith, 1947) was much too broad, encompassing several taxa superficially resembling this species. Maas Geesteranus (1986, 1992b) reported similar findings from his investigation of Smith's material.

The observation of latex and laticiferous hyphae in *M. elegantula* indicates that the infrageneric placement of the taxon must be reconsidered. Maas Geesteranus (1986, 1992b) placed the species in section Rubromarginatae Sing. ex Mass G., presumably due to the reddish lamellar edges and smooth to coarsely diverticulate cheilocystidia. However, none of the species within section Rubromarginatae are characterized by the presence of latex. Maas Geesteranus (1988, 1992b) segregates those taxa containing deeply colored latex into three sections: Sanguinolentae Maas G., Galactopoda (Earle) Maas G., and Crocatae Maas G. The placement of *M. californiensis* within section Galactopoda can be eliminated since all species of the section are characterized by smooth stipe cortical hyphae, and by stipes that turn black when specimens are dried, features not observed in *M. californiensis*. Of the remaining sections, the monospecific section Crocatae may also be eliminated due to the lack of marginate lamellae, pale stipe coloration, and clavate cheilocystidia with short, evenly spaced, cylindrical spinulae. Section Sanguinolentae is by far the most accommodating section for *M. californiensis*. The members of section Sanguinolentae are characterized macroscopically by reddish brown pileus and stipe coloration, red brown to purple brown lamellar edges, and dull orange to reddish brown latex. Microscopically the members of the section are characterized by cheilocystidia with reddish brown contents forming a sterile lamellar edge and occasionally covered with excrescences, smooth amyloid spores, and various other characters shared with *M. californiensis* (Maas Geesteranus, 1988, 1992b). The placement of *M. californiensis* in section Sanguinolentae is therefore proposed.

In addition to *M. californiensis*, the northern temperate members of section Sanguinolentae include *M. atkinsoniana* A.H. Sm. and *M. sanguinolenta*. To aid in distinguishing northern temperate *Mycena* species with deeply colored latex, the following artificial key is provided.

Key to Northern Temperate species of *Mycena* with deeply colored latex.

The following key is adapted from Mass Geesteranus (1988, 1992b), Smith (1947) and our own observations.

1. Stipe cortical hyphae smooth; stipe becoming black when dried ..sect. Galactopoda....*M. haematopus* (Pers.: Fr.) P. Kumm.
1. Stipe cortical hyphae diverticulate; stipe not becoming black when dried...2

2. Lamellar edges white, non-marginate; stipe apex white to orange; cheilocystidia clavate, covered with evenly spaced, short, cylindrical spinulae.....sect. Crocatae..... ....*M. crocata* (Schrad.: Fr.) P. Kumm.
2. Lamellar edges reddish brown to purplish brown; stipe apex light brown, brownish orange, reddish brown or purplish brown; cheilocystidia fusiform or irregularly clavate, smooth or with knob-like to long, cylindrical apical projections..........sect. Sanguinolentae...........3

3. Cheilocystidia contorted-clavate to irregular in shape, with irregular knob-like diverticula and/or numerous long, cylindrical apical projections.................... *M. californiensis*
3. Cheilocystidia fusiform to subcylindrical or rarely clavate, typically apically narrowed into one or more necks, acute, smooth or with occasional small diverticula......................4

4. Lamellae 23-26 reaching the stipe; basidiomes generally robust, stipe width 2-3 mm, pileus (5-) 10-30 mm broad .... *M. atkinsoniana*
4. Lamellae 13-21 reaching stipe; basidiomes generally smaller, stipe width 0.5-1.5 mm, pileus 7-18 mm broad .... *M. sanguinolenta*

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MYCOTAXON

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ADDENDUM TO ANNOTATED CHECK-LIST WITH HOST DATA AND DECAY CHARACTERISTICS FOR COLORADO WOOD ROTTING BASIDIOMYCOTINA

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ABSTRACT

Forty-two species of wood-rotting fungi are reported on new hosts in Colorado or as additional new records to the state. Twelve are brown-rot fungi, 26 are white-rot fungi, and four have unknown rot characteristics.

INTRODUCTION

The author published a checklist of Colorado wood-rotting fungi based on lists of collections from various herbaria (Lindsey 1988). Since then additional records have been collected by the author. Some of these are host records while others are new species records for the state. Several new hosts are included in this addendum; acronyms for these as well as other hosts given in this paper are listed below. Descriptions followed by a bold-face "**H**" are new host records.

Acronyms
ABBI = *Abies bicolor* A. Murry (corkbark fir)*
ABCO = *Abies concolor* (Gord. et Glend.) Lindl. ex Hildebr. (white fir)
ACSA = *Acer saccharinum* L (silver maple)
ARTR = *Artemisia tridentata* Nutt. (big sagebrush)
CEMO = *Cercocarpus montanus* Raf. (alder leaf mountain mahogany)
CHNA = *Chrysothamnus nauseosus* (Pallas) Britton (rabbitbrush)
FERU = *Fendlera rupicola* A.Gray (fendler bush)
PICO = *Pinus contorta* Dougl. ex Loud. (lodgepole pine)
PIED = *Pinus edulis* Engelm. (piñon pine)
PIEN = *Picea engelmannii* Parry ex Engelm. (engelmann spruce)
PIPO = *Pinus ponderosa* Laws. (ponderosa pine)
POAN = *Populus angustifolia* James (narrowleaf cottonwood)
ROPS = *Robinia pseudoacacia* L. (black locust)
SABA = *Salix babylonica* L. (weeping willow)
SYVU = *Syringa vulgaris* L. (lilac)
**Abies bicolor* was known in the previous checklist (Lindsey 1988) as *Abies lasiocarpa* v. *arizonica*.

CHECKLIST

Agaricales

Armillaria ostoyae (Romagn.) Herink; white root rot of living conifers and hardwoods, but also in dead trees and stumps; PIEN

Collybia acervata (Fr.:Fr.) P. Kumm; white rot of decaying conifer stumps or buried wood; PIEN

Coprinus atramentarius (Bull.:Fr.) Fr.; brown cubical butt and root rot of aspen; POTR

Hypholoma fasciculare (Huds.:Fr.) P. Kumm.; white rot of dead conifer logs and slash; PIEN; conifer chip mulch

Hypsizygus tessellatus (Bull.:Fr.) Singer; white rot of living or dead hardwoods; POAN

Lentinellus ursinus (Fr.) Kühner; white rot of conifer and hardwood logs and slash; PIEN

Megacollybia platyphylla (Fr.:Fr.) Kotl. & Pouzar; white rot of dead hardwood or conifer logs and slash; conifer **(H)**

Neolentinus lepideus (Fr.) Redhead & Ginns; brown cubical rot of dead trees and stumps, or dead wood in living trees; living POTR **(H)**

Neolentinus ponderosus (O.K. Mill.) Redhead & Ginns; brown cubical rot of dead trees and stumps; PIED **(H)**

Paxillus atrotomentosus (Batsch:Fr.) Fr.; brown rot of decaying conifer stumps, logs, or buried wood; rotted PIEN

Pholiota adiposa (Fr.:Fr.) P. Kumm.; white rot of dead conifers; ABCO **(H)**

Psathyrella candolleana (Fr.) A.H. Sm.; brown rot of well-decayed hardwood stumps or buried wood; ROPS **(H)**

Tricholomopsis rutilans (Fr.) Singer; white rot of decaying conifer wood; ABCO **(H)**

Coniophoraceae

Coniophora arida var. *arida* (Fr.) P. Karst.; brown cubical rot of dead conifer and hardwood; PIPO **(H)**

Coniophorella olivacea (Fr:Fr.) P. Karst.; brown cubical rot of conifer logs and slash; PIPO **(H)**

Corticiaceae

Acanthophysium cerrusatum (Bres.) Boidin; white rot of dead hardwood and conifer slash; ARTR, FERU, CEMO **(H)**

Amylocorticium cumminsii Gilb. & Lindsey; brown cubical rot of lodgepole pine in service; PICO timber in service

Athelia fibulata M.P. Christ.; rot not known; ARTR **(H)**

Botryobasidium ansosum (H.S. Jacks. & D.P. Rogers) Parm.; bark-inhabiting; PIEN, ABBI

Cristinia helvetica (Pers.) Parmasto; white rot of decayed hardwoods, occasionally conifers; CHNA

Hyphoderma medioburiense (Burt) Donk; white rot of well-decayed deciduous wood; POTR **(H)**

Hyphoderma subclavigerum (Bres.) Donk; white rot of conifer slash; PIEN

Hyphodontia arguta (Fr.) J. Erikss.; white rot of hardwood and conifer slash; PIED **(H)**

Hypochnicium bombycinum (Sommerf.:Fr.) J. Erikss.; white rot of hardwood and conifer slash; PIPO (H)

Leptosporomyces galzinii (Bourdot) Jülich; white rot of conifer slash, occasionally hardwood; PIPO (H)

Meruliopsis hirtellus (Burt) Ginns; White rot of twigs and branches; PIEN log, bark

Peniophora cinerea (Pers.:Fr.) Cooke; white rot of hardwood branches and slash; SYVU (H)

Peniophora pithya (Pers.) J. Erikss.; white rot of conifer logs and slash; down ABLA log

Phlebiella sulphurea (Pers.:Fr.) Ginns & Lefebvre; white rot of conifer and hardwood slash and bark; PIPO (H)

Tubulicrinus medius (Bourdot & Galzin) Oberw.; white rot of decorticated conifer branches, occasionally hardwoods; PIEN (H)

Tubulicrinus subulatus (Bourdot & Galzin) Donk; white rot of conifer slash, occasionally hardwood; PIPO (H)

Dacrymycetales

Dacrymyces chrysospermus (Schwein.) Berl & M.A. Curtis; brown rot; ABCO, POAN (H)

Ganodermataceae

Ganoderma applanatum (Pers.) Pat.; white root and butt rot of dead standing or living hardwoods, occasionally conifers; SABA, ABCO (H)

Hymenochaetaceae

Phellinus ferruginosus (Schrad.:Fr.) Bourdot & Galzin; white rot of dead hardwoods and conifers; ABBI

Polyporaceae

Antrodia radiculosa (Peck) Gilb. & Ryvarden; brown cubical rot of gymnosperms, rarely angiosperms; unknown conifer

Laetiporus sulphureus (Bull.:Fr.) Murrill; brown cubical butt rot of living conifers and hardwoods, but also found on stumps and dead standing trees; PIEN stump

Oxyporus corticola (Fr.) Ryvarden; white rot of dead conifer and hardwood; PIPO (H)

Trichaptum abietinum (Dicks.:Fr.) Ryvarden; white sapwood pocket rot of dead conifers; ABBI (H)

Sparassidaceae

Sparassis crispa (Wulfen:Fr.) Fr.; brown carbonizing root and butt rot of living conifers; living PIEN (H)

Thelephoraceae

Tomentella chlorina (Massee) G. Cunn.; rot not known; PIPO

Tomentella viridula Bourdot & Galzin; rot not known; CHNA

Tremellales

Exidia glandulosa (Bull.) Fr.; white saprot of hardwood slash and dead limbs on live trees; POAN (H)

ACKNOWLEDGEMENTS

The author thanks Donna Giersch for word processing this document and Dr. R.L. Gilbertson for helpful suggestions.

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FURTHER NEW SPECIES AND NEW REPORTS IN THE LICHEN FAMILY PARMELIACEAE (ASCOMYCOTINA) FROM SOUTH AFRICA

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ABSTRACT : The following new lichen species are described, *Canoparmelia terrapapia* Elix, *Flavoparmelia salazinica* Elix, *Hypotrachyna leeukopensis* Elix and *Paraparmelia maritima* Elix, and the new combinations *Canoparmelia rodriguesiana* (Müll. Arg.) Elix, *Karoowia lyrigera* (Brusse) Elix, *Neofuscelia nautilomontana* (Brusse) Elix and *Paraparmelia perfissa* (J.Steiner & Zahlbr.) Elix are made. *Canomaculina subcaperata* (Kremp.) Elix, *Hypotrachyna subpustulifera* Elix, *Parmotrema blanchetianum* (Müll. Arg.) Kalb and *Xanthoparmelia cravenii* Elix & J.Johnst. are reported for the first time from Africa, and *Relicina planiuscula* (Kurok.) Hale for the first time from South Africa.

Introduction

As a result of further study of the lichen family Parmeliaceae in South Africa (Elix 1997, Nash & Elix 1987), I am describing four new species. Throughout the present work chemical constituents were identified by thin layer chromatography (Culberson 1972; Culberson, Culberson & Johnson 1981; Culberson & Johnson 1982; Elix & Ernst-Russell 1993; Elix *et al.* 1988), high performance liquid chromatography (Feige *et al.* 1993) and comparison with authentic samples.

Canoparmelia terrapapia Elix sp. nov. Fig. 1

Species cum thallo ut in *Canoparmelia zimbabwensis* sed ab hac specie superficie epustulata differt.

Type: Republic of South Africa. Natal: 1 km NW of northern exit of Oribi Gorge Nature Reserve, paddock on plateau S of the road with single trees, 30°41'S, 30°16'E, *ca.* 480 m, on rocks and boulders, *D. Triebel & G. Rambold 7386*, 21 March 1990; holo: M; iso: CANB.

Thallus foliose, saxicolous, tightly adnate, to 2-4 cm wide. *Lobes* imbricate or not, crowded, ±plicate, sublinear-elongate, subdichotomously to irregularly branched, 0.5-1.2 mm wide, eciliate. *Upper surface* pale grey to grey, ±blackened at apices, shiny and smooth, becoming dull and rugulose, distinctly maculate, lacking pustules, soredia and isidia. Medulla white. Cell walls containing isolichenan. *Lower surface* smooth, jet black, brown at the lobe apices; rhizines moderately dense to dense, simple, black. *Apothecia* and *pycnidia* not seen.

Chemistry: Cortex K+ yellow; medulla K+ pale yellow-brown, C-, KC-, P+ red; containing atranorin (minor), chloroatranorin (minor), protocetraric acid (major), salazinic acid (trace), succinprotocetraric acid (trace), unknown fatty acid (trace).

In overall morphology this new species resembles *Canoparmelia zimbabwensis* (Hale) Elix & Hale, since both are characterized by tightly adnate, saxicolous thalli, crowded, irregular to sublinear lobes with a black lower surface and the presence of medullary protocetraric acid. However, *C. zimbabwensis* is clearly distinguished by producing laminal, erumpent pustules, whereas *C. terrapapia* lacks vegetative propagules. Morphologically this species could also be confused with the Australian *Paraparmelia rugulosa* Elix & J.Johnst., but the latter is clearly distinguished by the presence of *Xanthoparmelia*-type lichenan in the cell walls. At present *C. terrapapia* is only known from the type locality, adjacent to the Oribi Gorge Nature Reserve and its baboon (*Papio* sp.) colonies.

Specimens Examined

REPUBLIC OF SOUTH AFRICA. Natal, type locality, *D. Triebel & G. Rambold 7377, 7379*, 21 March 1990 (CANB, M - paratypes).

Flavoparmelia salazinica Elix, sp. nov. Fig. 2

Species cum thallo ut in *Flavoparmelia rutidota* sed ab hac specie sporis magis et acido salazinico continente differt.

Type: Republic of South Africa. Cape Province: Olifantskop, 25 km N of Paterson on E side of Highway R32, Grid 3325 BB, 650 m, on wood in scrub forest, *M. E. Hale 77565*, 16 Oct. 1986; holo: US; iso: CANB.

Thallus corticolous, foliose, adnate, firm and leathery, to 4–10 cm wide. *Lobes* imbricate, irregular, 2–5 mm wide; margins crenulate; lobulate or not;

apices subrotund. *Upper surface* yellowish green to green, ±rugulose at margins becoming very densely rugose towards the centre, dull or sometimes slightly shiny, lacking soredia, dactyls and isidia. *Medulla* white. *Lower surface* rugulose, black, with a narrow, brown marginal zone; sparsely rhizinate, the rhizines black, very coarse, simple to sparsely branched, 1–2 mm long. *Apothecia* sessile, 2-5 mm wide; disc concave then undulate-distorted, cinnamon-brown to dark brown; thalline exciple strongly involute at first, becoming rugulose and cracked. Ascospores ellipsoidal, 19-22 x 10-12 µm. *Pycnidia* not seen.

Chemistry. Cortex K-, medulla K+ yellow then dark red, C-, KC-, P+ orange-red; containing usnic acid (major), salazinic acid (major), consalazinic acid (minor), galbinic acid (minor), protocetraric acid (minor), norstictic acid (trace), secalonic acid A (trace).

Morphologically *F. salazinica* closely resembles *F. rutidota* (J.D. Hook. & Taylor) Hale, as these two species have similar adnate thalli with irregular, imbricate, rugose lobes, which lack vegetative propagules. However, they are clearly separated by their chemical constituents since salazinic acid, consalazinic acid and galbinic acid occur in *F. salazinica*, while *F. rutidota* contains protocetraric acid and caperatic acid. *F. rutidota* also differs in having significantly smaller spores (12-16 x 8-11 µm). These two species also exhibit very distinctive geographic distributions, with *F. rutidota* occurring in Australia and southern USA, whereas the present new species is known only from the type locality in Cape Province, South Africa.

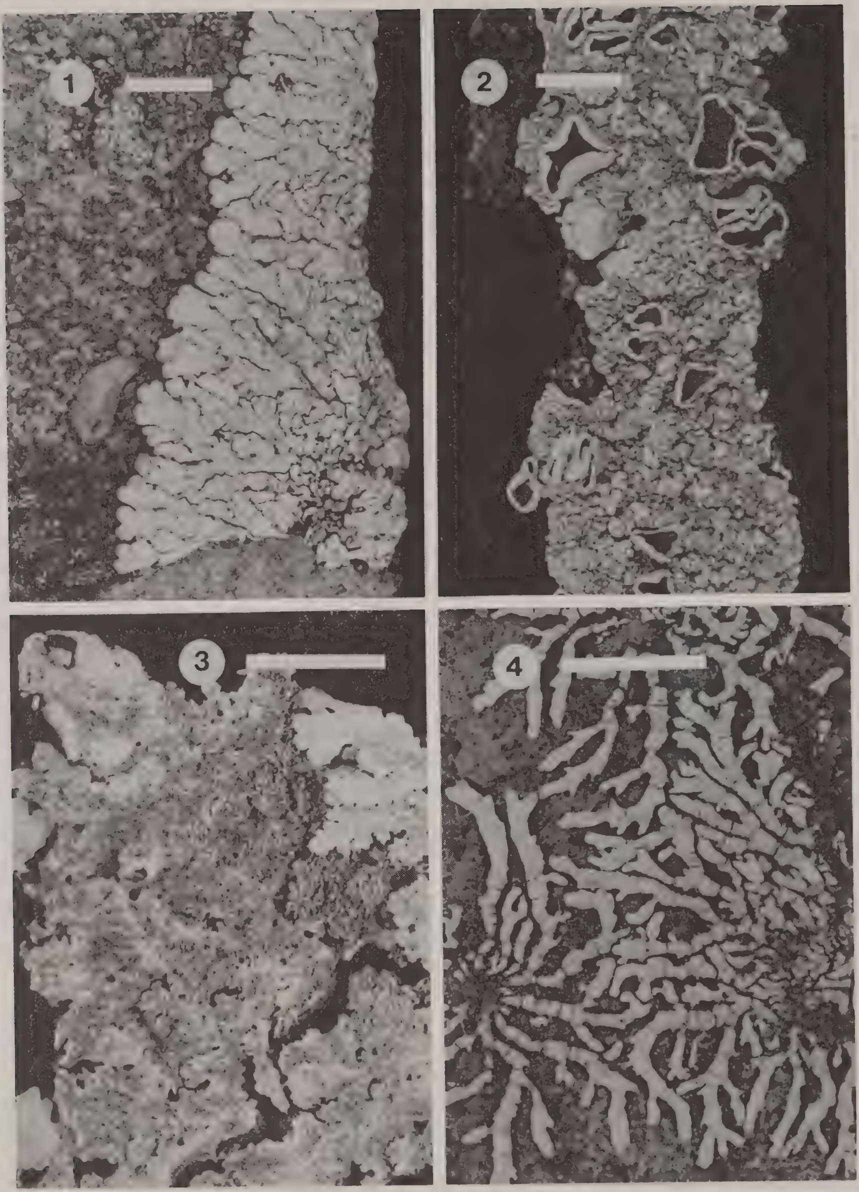
Hypotrachyna leeukopensis Elix, sp. nov. Fig. 3

Species cum thallo ut in *Hypotrachyna imbricatula* sed ab hac specie acido lobarico et acido protocetrarico continente differt.

Type: Republic of South Africa. Cape Province: Leeukop (Lion's Head), W of Cape Town, trail to summit, 33°56'S, 18°23'E, 600 m, on E-exposed rocks, *D. Triebel & G. Rambold 8376*, 5 April 1990; holo: M; iso: CANB.

Thallus foliose, loosely adnate, saxicolous, to 5 cm wide. *Lobes* often crowded, imbricate, sublinear to subirregular, irregularly branched, 1.5-4.5 mm wide. *Upper surface* grey but darkening with age, shiny at the tips, soon becoming white-maculate, isidiate, lacking soredia and pustules; isidia moderate to dense, cylindrical, simple, slender, apices syncorticate and darkening. Medulla white. *Lower surface* black, brown at the tips; rhizines dense, dichotomously branched. *Pycnidia* and *apothecia* not seen.

Chemistry. Cortex K+ yellow; medulla K+ pale yellow-brown, C-, P+ red; containing atranorin (major), chloroatranorin (major), usnic acid (minor), lobaric acid (major), protocetraric acid (major), norstictic acid (minor), unknown fatty acid (minor).



Figures 1-4. New species of Parmeliaceae: 1, *Canoparmelia terrapapia* (holotype in M); 2, *Flavoparmelia salazinica* (isotype in CANB); 3, *Hypotrachyna leeukopensis* (holotype in M); 4, *Paraparmelia maritima* (holotype in M). Scale bar = 5 mm.

Morphologically this species resembles *Hypotrachyna imbricatula* (Zahlbr.) Hale in having loosely adnate, fragile thalli with imbricate, sublinear to subirregular lobes and cylindrical isidia, but has quite distinctive medullary chemistry. Whereas *H. leeukopensis* produces protocetraric acid and lobaric acid, *H. imbricatula* contains the barbatic acid complex in the medulla. Indeed, both the medullary and cortical chemistry observed in *H. leeukopensis* are unique for this genus, although other species do contain protocetraric acid and lobaric acid, albeit not jointly. For instance, the Asian species *Hypotrachyna koyaensis* (Asah.) Hale, contains atranorin, chloroatranorin, protocetraric acid (major) and virensic acid (trace), but lacks the other constituents observed in *H. leeukopensis.* Further, *H. koyaensis* has broader (3-10 *cf.* 1.5-4.5 mm wide), apically rotund, moderately rhizinate lobes and ±coralloid branched isidia. At present this species is only known from the type collection.

Paraparmelia maritima Elix, sp. nov. Fig. 4

Species cum thallo ut in *Paraparmelia aranaea* sed ab hac specie acido fumarprotocetrarico continente differt.

Type: Republic of South Africa. Cape Province: Tsitsikamma Coastal National Park, unnamed hill N of township, Nature's Valley trail to Kalanderpad Lookout, 33°58'S, 23°33'E, *ca.* 200 m, on small stones, *D. Triebel & G. Rambold 7659*, 28 March 1990; holo: M.

Thallus small-foliose to subcrustose, tightly adnate, saxicolous, forming irregular rosettes, 1-5 cm wide. *Lobes* scarcely imbricate, separate at apices, ±contiguous towards thallus centre, sublinear to sublinear-elongate, dichotomously branched, 0.2-1.0 mm wide; broader at apices. *Upper surface* pale grey to dark grey, flat, becoming dull, smooth, older lobes becoming tangentially cracked and areolate in thallus centre; lacking soredia and isidia. *Medulla* white. *Lower surface* black, flat or rugulose; rhizines very sparse, simple, black; for the most part lower surface attached directly to the substratum. *Apothecia* and *pycnidia* not seen.

Chemistry. Cortex K+ yellow; medulla K-, C-, KC-, P+ brick red; containing atranorin (minor), fumarprotocetraric acid (major), quaesetic acid (minor), galbinic acid (minor), protocetraric acid (minor), salazinic acid (trace).

This species is characterized by the tightly adnate thallus, the narrow, separate, linear-elongate lobes with a black lower surface, the lack of isidia and the presence of fumarprotocetraric acid in the medulla. Morphologically it resembles the Australian species *Paraparmelia aranaea* Elix & J.Johnst., but the latter differs in having slightly broader lobes (0.2-1.2 mm wide) and more particularly, in containing protocetraric acid in the medulla. At present this new species is known only from the type locality, where it appears to be quite common.

Specimens Examined

REPUBLIC OF SOUTH AFRICA. Cape Province, type locality, *D. Triebel & G. Rambold 7652, 7656, 7657, 7660, 7662, 7668,* 28 March 1990 (ASU, CANB, M - paratypes).

New Combinations

Canoparmelia rodriguesiana (Hue) Elix, comb. nov.
Basionym: *Parmelia rodriguesiana* Hue, *Nouv. Arch. Mus. Paris, ser.* 3, **1**: 167 (1899).

Karoowia lyrigera (Brusse) Elix, comb. nov.
Basionym: *Parmelia lyrigera* Brusse, *Mycotaxon* **35**: 24 (1989).

Neofuscelia nautilomontana (Brusse) Elix, comb. nov.
Basionym: *Parmelia nautilomontana* Brusse, *Mycotaxon* **49**: 7 (1993).

Paraparmelia perfissa (Steiner & Zahlbr.) Elix, comb. nov.
Basionym: *Parmelia perfissa* Steiner & Zahlbr., *Bot. Jb.* **60**: 519 (1926).

New Records for Africa

Canomaculina subcaperata (Kremp.) Elix, *Mycotaxon* **65**: 477 (1997).

Specimens Examined

REPUBLIC OF SOUTH AFRICA. Orange Free State, Magaliesberge, W of Pretoria, Breedtsnek, saddle 70 m W and below summit, 25°52'S, 27°27'E, *ca.* 1700 m, on shaded rocks, *D. Triebel & G. Rambold 8663,* 13 April 1990 (CANB, M).
ZIMBABWE. Eastern Highlands, Stanhope Block, 18°11'S, 31°46'E, 2240 m, on granite, *U. Becker 251089, 251113C,* 23 March 1994 (KOELN).

Hypotrachyna subpustulifera Elix, *Mycotaxon* **47**: 108 (1993).

Specimen Examined

REPUBLIC OF SOUTH AFRICA. Natal, Royal National Park, in narrow gorge "The Crack", W of Plowman's Kop, 28°41'S, 28°55'E, *ca.* 1900-1950 m, on wet S-exposed boulders, *D. Triebel & G. Rambold 6776*, 10 March 1990 (M).

Parmotrema blanchetianum (Müll. Arg.) Kalb, *Lichenes Neotropici* Fasc. 9, no. 392 (1986).

Specimens Examined

REPUBLIC OF SOUTH AFRICA. Transvaal, Mac Mac Falls, 13 km from Sabie, 24°59'30"S, 30°48'E, *ca.* 1400 m, on sandstone, *T. H. Nash 23452*, 16 Jan. 1986 (ASU, CANB).
ZIMBABWE. Eastern Highlands, Stanhope Block, 18°11'S, 31°46'E, 2240 m, on granite, *U. Becker 251121*, 23 March 1994 (KOELN).

Xanthoparmelia cravenii Elix & J.Johnst., *Bull. Brit. Mus. (Nat. Hist.), Bot.* **15**: 223 (1986).

Specimen Examined

REPUBLIC OF SOUTH AFRICA. Cape Province, near Huisrivier Pass, near Ladismith-Calitzdorp road, 2 km E of Gamkarrivier, 2 km along road to Matjiesvlei, 33°28'S, 21°32'E, *ca.* 400 m, on pebbles and small boulders, *D. Triebel & G. Rambold 7796*, 29 March 1990 (M).

New Record for South Africa

Relicina planiuscula (Kurok.) Hale, *Phytologia* **28**: 484 (1974).

Specimens Examined

REPUBLIC OF SOUTH AFRICA. Natal, Oribi Gorge Nature Reserve, Baboons View, NW of Hutted Camp, 30°44'S, 30°16'E, *ca.* 450 m, on bark, *D. Triebel & G. Rambold 7298, 7306*, 21 March 1990 (M).

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I thank Uwe Becker (KOELN), Drs Dagmar Triebel and Gerhard Rambold (both M), Professor Thomas H Nash III (ASU) and the late Dr Mason E Hale (US) for generously making their excellent collections of Parmeliaceae from southern Africa available to me. I also wish to thank Stuart Hay and Neal McCracken of the Photographic Unit, ANU, for preparing the photographs, Doug Verdon for checking the Latin descriptions and the following herbaria for the loan of critical collections: BOL, LD, PRE, PREM, US.

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Aphyllophorales (Basidiomycotina) of arid habitats of the Canary Islands. Preliminary data.

by

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ABSTRACT

Thirty-one species of Aphyllophorales, which appear occasionally in the desert-arid-semiarid areas of the Canary Islands have been studied. All of them are recorded as new for these habitats, though most are known from forest ecosystems. Eighteen species are new to the particular islands (6 each for Lanzarote and Fuerteventura, 1 for Gran Canaria, 3 for Tenerife, and 1 for Gomera and La Palma, respectively). *Acanthophysium minor* (Pilát) Tellería, *Oidium candicans* (Sacc.) Linder, *Peniophora nuda* (Fr.) Bresad. and *Phanerochaete xerophila* Burds., are new to the Canarian fungi checklist.

Key words: Aphyllophorales, Canary Islands, arid-semiarid areas.

INTRODUCTION

Up to now, studies on Aphyllophorales of the Canary Islands have mainly been carred out in the forest areas located at the mesomediterranean and thermomediterranean subhumid-humid bioclimatic belts, i.e. in pine forests and evergreen forest (laursilva and *Erica-Myrica* forest), respectively.

In this paper, preliminary results are reported for this fungal group for the first time in arid-semiarid-dry zones of the islands are given.

According to Rivas-Martínez *et al.* (1993), the Canary bioclimate is Mediterranean in type, represented by three subtypes: 1) Mediterranean-desertic; 2) Mediterranean xerophytic and 3) Mediterranean mesophytic, with arid ombrotypes (100-200 mm precipitation), semi-arid (200-300 mm), dry (350-550 mm), subhumid (550-800 mm), humid (> 800 mm), respectively. In the Canary Islands are represented most of bioclimatic Mediterranean thermotypes

The investigated localities are situated altitudinally from the inframediterranean desertic and arid ombrotype to the thermomediterranean semiarid-dry xerophytic bioclimatic belts, and from the higher vegetation point of view corresponding to: a) succulent scrublands of *Euphorbia* spp. shrub communities; b) fruticedes and xerophytic groves (*Juniperus turbinata* subsp. *canariensis, Olea europaea* subsp. *cerasiformis* and *Pistacia atlantica* communities); and c) open substitution scrublands belonging of the vegetation of semiarid, dry and occasionally sub-humid xerophytic infra-thermomediterranean belts (*Cistus* and *Micromeria* spp. communities). In the study area edaphohydrophilic communities were also found: a) *Tamarix* edapho-hidro-halophilous community of the desert-xerophytic inframediterranean belt; and b) azone *Salix* edapho-hidrophilic mesophytic community distributed from the infra to supramediterranean belts.

MATERIALS AND METHODS

Collections were made in nearly all of the islands [Lanzarote (L), Fuertevetura (F), Gran Canaria (C), Tenerife (T), Gomera (G), La Palma (P)]. In field work, care was taken to record as much ecological data regarding altitude, exposure, etc. of the biotope along with higher vegetation, substrate, etc., associated with the fungal species. Material was identified in the laboratory, following standard taxonomic methodology. All the exsiccata are deposited in the Herbarium TFC, Mycological Section (here called TFC Mic.) of the University of La Laguna. Additionally making provisional ecological evaluations on Aphyllophorales of the desertic-arid-semiarid-dry zones of the Canaries in relation to these in habitats more favorable to fungal development. The illustrations were made with the aid of a drawing tube and the exsiccatum number on which was made the drawings is indicated in each case.

INVESTIGATED LOCALITIES

Lanzarote:

(1) Haría: Barranco de Temisa, 615 m a.s.l.; UTM: 28RFT 449228; Orientation NE. Degraded xerophytic brushwood of *Euphorbia* spp. community.

(2) Near Haría, 450 m a.s.l.; 28RFT 448235; N. Plantations of *Pinus halepensis* Mill. with xerophitic elements of *Euphorbia* spp. communities, in a very degraded zone.

(3) Haría: 300 m a.s.l.; 28RFT 457243; NW. *Tamarix* community along the road.

(4) Haría: Arrieta, Punta Mujeres, 25 m a.s.l.; 28RFT 515255; E. Excelent *Euphorbia balsamifera* communty.

(5) Teguise: Vega de Tahiche, 150 m a.s.l.; 28RFT 420115; E. Degraded and xerophytic brushwood of *Launaea arborescens* (Batt.) Murb., *Tamarix canariensis*, etc., community, in old cultivated areas today abandoned.

Fuerteventura:

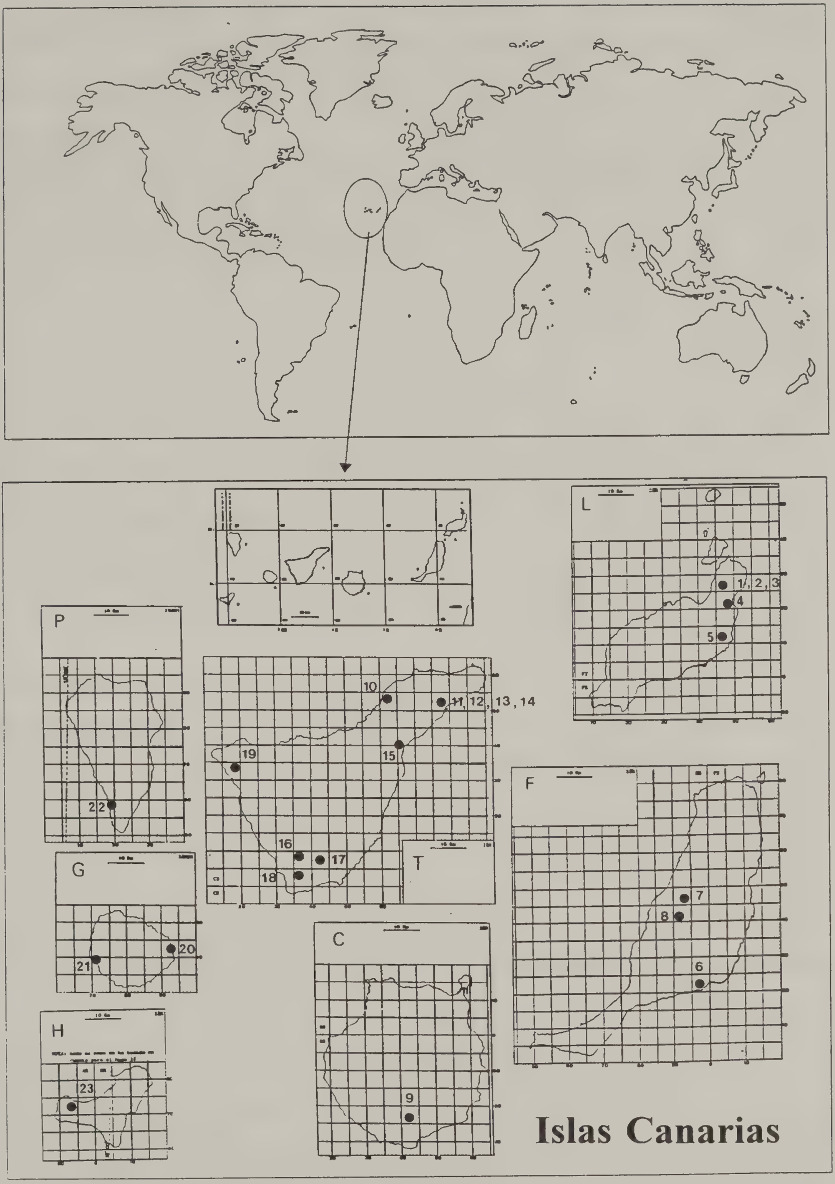
(6) Tuineje: Gran Tarajal, 0 m a.s.l.; 28RES 967208; S. Edapho-hidro-halophilic *Tamarix canariensis* Willd., *Phoenix canariensis* Chab., and *Suaedea vera* Forssk. *ex* J.F. Gmel. community, in the mouth of the watercourse at the sea level.

(7) Betancuria: 450 m a.s.l.; 28RES 930431; NW. Plantations of *Pinus canariensis* Chr. Sm. *ex* DC. *in* Buch and *Eucalyptus* sp., mixed with xerophytic elements of the semiarid-dry infra-thermomediterranean bioclimatic belts communities;

(8) Betancuria: Vega de Río Palmas, Presa de la Peñita, 215 m a.s.l.; 28RES 891407; SW. Edapho-hydro-halophilic community of *Tamarix canariensis* Willd. and *Phoenix canariensis* Chab., on sandy soil of the desert-xerophitic community of inframediterranean bioclimatic belt.

Gran Canaria:

(9) San Bartolomé de Tirajana: Barranco de La Data, 200 m a.s.l.; 28RDR 397739; S. Succulent scrublands of *Euphorbia* spp. communities, with scattered palms trees.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Localities map

Tenerife:

(10) Tacoronte: cliff of the Bahía de la Garañona, 200 m a.s.l.; 28RCS 598515; N. Succulent scrublands of *Euphorbia* spp. community of the inframediterranean bioclimatic belt.

(11) Santa Cruz de Tenerife (Anaga): Barranco de Crispín, 150 m a.s.l.; 28RCS 795547; E. Succulent scrublands of *Euphorbia* spp. community of the inframediterranean bioclimatic belt.

(12) *Ibid*., Anaga: Barranco de Salazar, 200 m a.s.l.; 28RCS 809533; E. Succulent scrublands of *Euphorbia* community mixed with aisolet palm trees; and occasionally the vegetation is representated by a humide *Rubus inermis-Salix canariensis* community; in the inframediterranean bioclimatic belt.

(13) *Ibid*., Anaga: Barranc. de San Andrés, 150 m a.s.l.; 28RCS 823562; E. Succulent scrublands of *Euphorbia* spp. community, mixed with palm trees aisolet, in the inframediterranean bioclimatic belt.

(14) *Ibid*., Anaga: Valle Brosque, near San Andrés, 300 m a.s.l.; 28RCS 801558; E. Succulent scrublands of *Euphorbia* community mixed with aisolet palm trees; and occasionally the vegetation is representated by a humide *Rubus inermis-Salix canariensis* community; in the inframediterranean bioclimatic belt.

(15*) Candelaria: Barranc. Hondo, 100 m a.s.l.; 28RCS 689420; E.

(16) Vilaflor: Mirador de Jama, 900 m a.s.l.; 28RCS 376109; S. *Euphorbia* spp. communities very degraded, with *Ficus carica* L., *Opuntia ficus-indica* (L.) Mill., *Pistacia atlantica* Desf., *Cistus monspeliensis* L., etc., in the thermomediterranean dry-subhumed.

(17) San Miguel de Abona: Las Cuevitas, 650 m a.s.l.; 28RCS 405097; S. *Euphorbia* spp. community very degraded of the thermomediterranean bioclimatic belt.

(18) Arona: Valle de San Lorenzo, Las Toscas, 400 m a.s.l.; 28RCS 373089; S. in *Cistus monspeliensis-Micromeria hyssopifolia* community, installed in abandoned cultivation terrace.

(19) Santiago del Teide: Masca, Risco Tarucho, 975 m a.s.l.; 28RCS 191235; SW. *Euphorbia atropurpurea* (Brouss.) Webb & Berth. and *Retama raetam* (Forssk.) Webb & Berth communit, of the thermomediterranean bioclimatic belt.

Gomera:

(20) San Sebastián.: Playa de Avalo, 0 m a.s.l.; 28RBS 934112; E. *Tamarix canariensis* community at a ravine mouth in non halophilic soil, of the inframediterranean bioclimatic belt.

(21) Valle Gran Rey: 10 m a.s.l.; 28RBS 705097; W. Degraded *Euphorbia* spp. communities with palm trees, of the inframediterranean bioclimatic belt.

La Palma:

(22) Fuencaliente: near lava flows of El Charco, 800 m a.s.l.; 28RBS 198598; W. Anthropic and edaphoxerophytic pine forest, with elements in the undergrowth of *Euphorbia* spp. communities in the thermomediterranean bioclimatic belt.

Hierro

(23*) Frontera: Sabinar de la Dehesa, 570 m a.s.l.; 28RAR 921733; W

Remark: For its nearness, the localities numbers 1, 2, 3, of Lanzarote and 11, 12, 13, 14 of Tenerife, are represented on the map by only one point, respectively. (*)Bibliographic records.

RESULTS

Thirty-one species of corticicolous Aphyllophorales have been recorded occuring occasionally in the desertic-xerophytic areas of the Canary Islands. All of them are new for these habitats, though most are known from forest ecosystems, where they have their optimal frequency (Rodríguez-Armas & Beltrán Tejera, 1995). Eighteen species are new to the particular islands (6 each for Lanzarote and Fuerteventura, 1 for Gran Canaria, 3 for Tenerife, and 1 for Gomera and La Palma, respectively).

Acanthophysium minor (Pilát) Tellería, *Oidium candicans* (Sacc.) Linder, *Peniophora nuda* (Fr.) Bresad. and *Phanerochaete xerophila* Burds., are new to the Canarian Islands. Although uniform seasonal monitoring was not performed, most of the species appear in the autumn and winter months from November to January, exceptionally in April due to occasional rain. Normally these localities are extremely dry and drought in the last few winters is probably the reason for the small number of species. The highest biodiversity of especies is in the *Corticiaceae* (24 species).

Annotated list of the species studied:

Corticiaceae:

Acanthophysium minor (Pilát) Tellería, *Nova Hedwigia* 53: 231. 1991.
(Aleurodiscus cerussatum (Bresad.) Höhnel & Litsch. var. *minor* Pilát)
HABITAT AND DISTRIBUTION INSULAR.- **Tenerife (19)**, on decayed branches of *Euphorbia obtusifolia* Poir.; 28.XII.1994 (TFC Mic. 6976: Fig. 1).
REMARKS.- There has been a controversy around this taxon probably since the Bourdot & Galzin's (1912: 351) record of specimens very close to *Aleurodiscus cerussatus* (Bres.) Höhnel & Litsch., which has smaller spores than Bresadola's taxon (*Corticiun cerussatum* Bresad., 1892: 37). Later Pilát described the variety *minor* which included the smaller spores complex. This problem has been dealt with by Tellería (1991a), who accepted *Acanthophysium minor* (Pilát) Tellería, with *Aleurodiscus cerussatus* (Bresad.) Höhnel & Lisch. var. *minor* Pilát as basionym. We think the Canarian material should be called *A.minor* (Pilát) Tellería, though the spores can 12.3 µm be up long. Boidin & Gilles (1986: 295), agree with Lemke (1964 *in op.cit.*), when they say that *Acanthophysium cerussatum* (Bres.) Boid., is a collective species and ... "l'existence et les limites des différentes espèces comprises sous ce nom nécessiteraient la culture, c'est-à-dire les tests d'intercompatibilité". This taxon has been recorded from several localities of the Iberian Peninsula (Tellería, 1991b); and recently Melo (1994), has mentioned it for Portugal, on dry stems of *Cytisus* sp., at 550 m a.s.l. It is a new record for the Canary Islands.

Amphinema byssoides (Pers.: Fr.) John Erikss., *Symb. Bot. Upsal.* 16 (1): 112, 1958.
HABITAT AND DISTRIBUTION INSULAR.- **Lanzarote (2)**, on fallen branch of *Pinus halepensis*; 20.II.1993 (TFC Mic. 6927).
REMARKS.- It is new for Lanzarote but previously mentioned for several islands (C, T, G, P), in laurisilva, pine forest and pine forest mixed with *Erica-Myrica* community.

Byssomerulius corium (Pers.: Fr.) Parmasto, *Izv. Akad. Nauk Estonsk. KKR, Ser. Biol.* 16 (4): 383, 1967.
HABITAT AND DISTRIBUTION INSULAR.- **La Palma (22)**, on fallen branches of *Euphorbia obtusifolia* Poir.; 3.XI.1990, 28.VI.1991, 8.XI.1991 (TFC Mic. 6198, 6387, 6401).
REMARKS.- It is new for La Palma, but has early been recorded for the Canary Islands (C, T, G, H, P), in evergreen forest, pine forest and pine forest mixed with *Myrica-Erica* community; and in mesophitic populations of introduced species (*Quercus robur* L., *Castanea sativa* Mill., etc.), (Beltrán & Rodríguez-Armas, 1993).

Ceraceomyces tessulatus (Cooke) Jülich, *Willdenowia Beih.* 7: 154. 1972.
HABITAT AND DISTRIBUTION INSULAR.- **Fuerteventura (6)**, on dead leaves of *Phoenix canariensis*; 5.XII.1994 (TFC Mic. 7129, 7130).
REMARKS.- New for Fuerteventura; it has been noted from other islands (G, P), in evergreen forest.

Globulicium hiemale (Laurilia) Hjortstam, *Svensk Bot. Tidskr.* 67 (2): 108, 1973.
HABITAT AND DISTRIBUTION INSULAR.- **Gran Canaria (9)**, on dead leaves of *Phoenix canariensis*; 26.V.1991 (TFC Mic. 7139). **Tenerife (12)**, on the same substrat; 2.IV.1993 (TFC Mic. 6603).

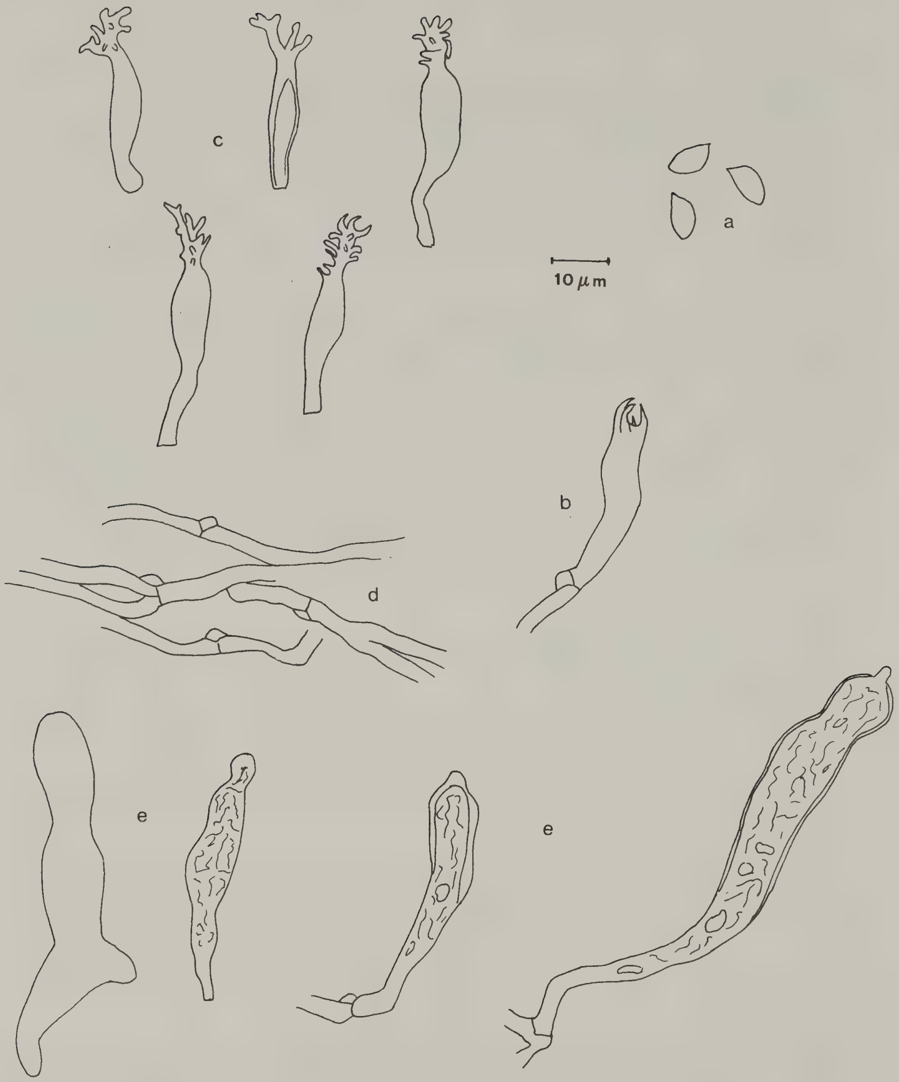


Fig. 1.- ***Acanthophysium minor*** (Pilát) Tellería. A.Spores; b.Basidia; c.Dendrohyphidia; c. Generative hyphae; e.Gloeocystidia.

REMARKS.- A new for Gran Canaria and Tenerife. Previously noted from other islands (T, G, H, P), in evergreen forest and pine forest mixed with *Erica-Myrica* community.

Hyphoderma medioburiense (Burt) Donk, *Fungus* 27 (1-4): 15, 1957.
HABITAT AND DISTRIBUTION INSULAR.- **Fuerteventura (7)**, on dead trunk of *Eucalyptus* sp.; 4.XII.1994 (TFC Mic. 6920, 7132). **Tenerife (10)**, on unidentified branches. ***Ibid.***, 300 m a.s.l., on debris of *Rumex lunaria* L.; 14.XI.1994 (TFC Mic. 6890, 6888). **Tenerife (17)**, on debris of *Kleinia neriifolia* Haw; 13.XI.1994 (TFC Mic. 6947). **Tenerife (19)**, on the same substrate; 28.XII.1994 (TFC Mic. 7148, 7147).
REMARKS.- It is a new record for Fuerteventura; noted early from Tenerife in evergreen forest.

Hyphoderma praetermissum (P.Karst.) John Erikss. & Strid., *in* John Eriksson & Ryvarden, *Cortic. North Europe* 3: 505, 1975.
HABITAT AND DISTRIBUTION INSULAR.- **Fuerteventura (7)**, on decaying wood of *Pinus canarienis*; 4.XII.1994 (TFC Mic. 7110). **Tenerife (11)**, on dead branches of *Euphorbia obtusifolia*; 5.XI.1994 (TFC Mic. 6937). **Tenerife (19)**, on debris of the same substrate; 28.XII.1994 (TFC Mic. 6969, 7133). **La Palma (22)**, on dead branches of the same substrate; 3.XI.1990 (TFC Mic. 6201).
REMARKS.- It is a new record for Fuerteventura, but has been noted from other islands (C, T, G, P), in forest communities.

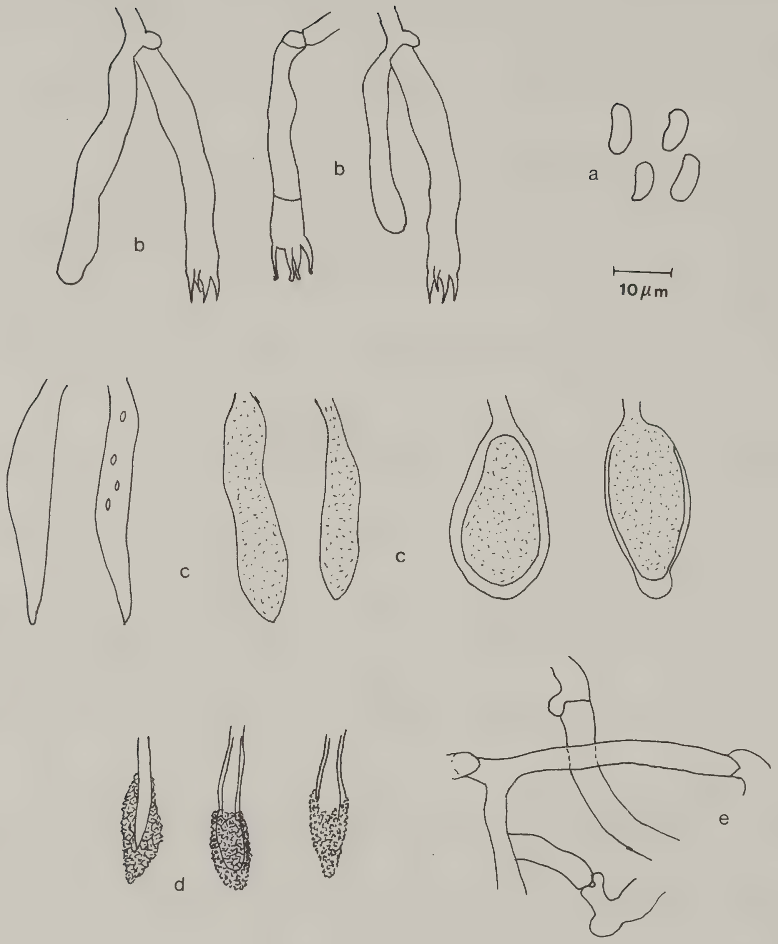
Hyphoderma setigerum (Fr.) Donk, *Fungus* 27 (1-4): 15, 1957.
HABITAT AND DISTRIBUTION INSULAR.- **Tenerife (12)**, on dead leaves of *Phoenix canariensis*; 2.IV.1993 (TFC Mic. 6601).
REMARKS.- It is new for Tenerife, but previously known in the laurel forest of La Palma.

Hyphodontia sambuci (Pers.) John Erikss., *Symb. Bot. Upsal.* 16 (1): 104, 1958.
HABITAT AND DISTRIBUTION INSULAR.- **Lanzarote (1)**, on fallen branch of *Nauplius intermedius* Webb; 16.I.1994 (TFC Mic. 7096, 7097). **Tenerife (12)**, on unidentified debris; 2.IV.1993 (TFC Mic. 6600). **Tenerife (16)**, on debris of *Opuntia ficus-indica*; 20.III.1994 (TFC Mic. 6409).
REMARKS.- It is new for Lanzarote, but previously noted in Tenerife and La Palma, in forest communities.

Oidium candicans (Sacc.) Linder
HABITAT AND DISTRIBUTION INSULAR.- **Fuerteventura (7)**, on decaying wood of *Eucalyptus* sp.; 4.XII.1994 (TFC Mic. 7111).
REMARKS.- *Botryobasidium candicans* John Erikss., han been reported from the Canary Islands (T, G, P), but not in the conidial state.

Peniophora boidinii Reid., *Rev. Biol. Lisboa* 5: 146, 1965.
HABITAT AND DISTRIBUTION INSULAR.- **Tenerife (17)**, on fallen branch of *Ficus carica*; 13.XI.1994 (TFC Mic. 6964).
REMARKS.- Noted early from a very humid laurel forest in La Palma, we now report this new collection in another island and habitat. According to Boidin (1994), this species is known from Mediterranean to the Macaronesian islands (Azores, including our first report to the Canary Islands in Rodríguez-Armas *et al.*, 1992).

Peniophora cinerea (Pers.: Fr.) Cooke, *Grevillea* 8 (45): 20, 1879.


b
b
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c
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e

Fig. 2.- ***Peniophora nuda*** (Fr.) Bresad. a. Spores; b. Basidia; c. Sulfocystidia; d. Incrusted cystidia; e. Generative hyphae.

HABITAT AND DISTRIBUTION INSULAR.- **La Palma (22)**, on dead branches of *Euphorbia obtusifolia*; 28.VI.1991 (TFC Mic. 6781).
REMARKS.- Common in the Canary Islands (C, T, G, H, P), in evergreen forest, pine forest and pine forest mixed with *Erica-Myrica* community.

Peniophora incarnata (Pers.: Fr.) P.Karst., *Hedwigia* 28 (1): 27, 1889.
HABITAT AND DISTRIBUTION INSULAR.- **Fuerteventura (7)**, on decaying bark of *Pinus canariensis*; 4.XII.1994 (TFC Mic. 7131).
REMARKS.- Previously known in the Canary Islands (C, T, G, P), in evergreen and pine forest. It is a new report for Fuerteventura.

Peniophora lycii (Pers.) Höhn. & Litsch., *Sitzungsber. Kaiserl. Akad. Wiss. Math.-Naturwiss. Cl. Abt.* 1, 116: 747, 1907.
HABITAT AND DISTRIBUTION INSULAR.- **Lanzarote (1)**, on fallen branches of *Nauplius intermedius*; 16.I.1994 (TFC Mic. 7098, 7099, 7100). **Fuerteventura (7)**, on fallen branches of *Eucalyptus* sp. and *Launaea arborescens* (Batt.) Murb.; 4.XII.1994 (TFC Mic. 7105, 7106, 7107, 7108, 7109). **Tenerife (13)**, on dead leaves of *Phoenix canariensis*; 2.IV.1993 (TFC Mic. 6944). **Tenerife (12)**, on decayed branches of *Periploca laevigata* Ait.; 2.IV.1993 (TFC Mic. 6597, 6598). **Tenerife (14)**, on decayed branches of *Artemisia thuscula* Cav.; 5.XI.1994 (TFC Mic. 6936). **Tenerife (17)**, on fallen branches of *Ficus carica*; 13.XI.1994 (TFC Mic. 6963). **Gomera (21)**, on dead leaves of *Phoenix canariensis*; 4.III.1992 (TFC Mic. 6399). **La Palma (22)**, on dead branches of *Euphorbia obtusifolia*; 3.XI.1990, 8.XI.1991 (TFC Mic. 6182, 6429).
REMARKS.- Previously noted from the Canary Islands (C, T, G, H, P), in evergreen forest and pine forest mixed with *Erica-Myrica* community. New for Lanzarote and Fuerteventura, in a xerophytic area.

Peniophora meridionalis Boidin, *Bull. Soc. Mycol. France* 74 (4): 1958 [1959].
HABITAT AND DISTRIBUTION INSULAR.- **Lanzarote (4)**, on fallen branches of *Tamarix canariensis*; 21.II.1993 (TFC Mic. 6929). **Lanzarote (3)**, on the same substrate; 21.II.1993 (TFC Mic. 6928, 6930, 7086).
REMARKS.- Previously reported in the Canaries only in evergreen forest and pine forest mixed with *Erica-Myrica* community (C, T, G, H, P). New record for Lanzarote.

Peniophora nuda (Fr.) Bresad., *Atti Imp. Regia Accad. Rovereto, ser.* 3, 3 (1): 114, 1897.
HABITAT AND DISTRIBUTION INSULAR.- **Tenerife (13)**, on fallen leaves of *Phoenix canariensis*; 2.IV.1993 (TFC Mic. 6707: Fig. 2). **Tenerife (17)**, on fallen branches of *Ficus carica*; 13.XI.1994 (TFC Mic. 6958, 6960, 6965). **Tenerife (19)**, on fallen branches of *Kleinia neriifolia*; 28.XII.1994 (TFC Mic. 7146).
REMARKS.- No previous record is known for the Canary Islands.

Phanerochaete andreae Bursd., Beltrán-Tej. & Rodríguez-Armas, *Mycotaxon* 54: 296, 1995.
HABITAT AND DISTRIBUTION INSULAR.- **Lanzarote (1)**, on dead branches of *Nauplius intermedius*; 16.I.1994 (TFC Mic. 7102). **Lanzarote (5)**, on unrecognized decayed wood; 19.XII.1994 (TFC Mic. 7101).
REMARKS.- This taxon was described originally fromGomera: Parque Nacional de Garajonay, Cherelepín, 1200 m a.s.l., in *Erica-Myrica* community with *Cistus monspeliensis*, in the thermomediterranean mesophytic subhumid bioclimatic belt. The ocurrence of this species

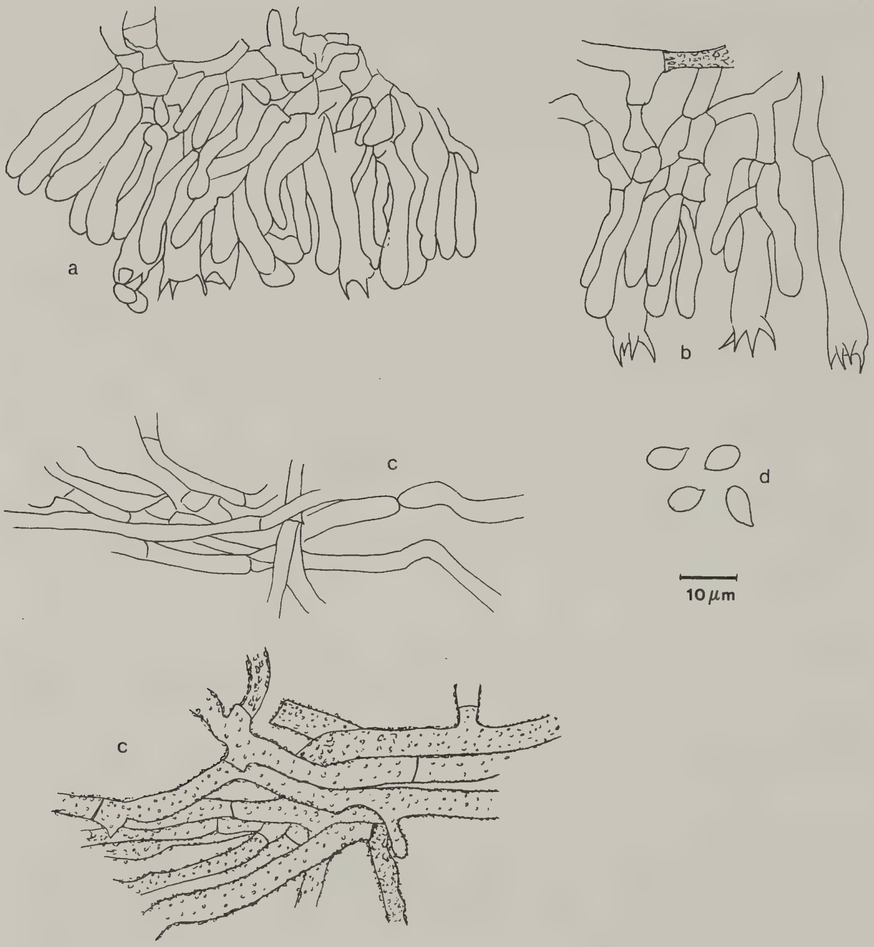

Fig. 3.- ***Phanerochaete xerofila*** Burs. a. Hymenial elements; b. Basidia; c. Generative hyphae and incrusted; d. Spores.

in the xerophytic arid-semiarid-dry inframediterranean bioclimatic belt of the Lanzarote confirms the wide ecological adaptability of a large number of Aphyllophorales at least in the Canary Islands.

Phanerochaete martelliana (Bres.) John Erikss. & Ryvarden *in* John Eriksson *et al., Cortic. North Europe* 5: 1011, 1978.
HABITAT AND DISTRIBUTION INSULAR.- **Tenerife (12)**, on decayed wood of *Periploca laevigata*; 2.IV.1993 (TFC Mic. 6596).
REMARKS.- It is recorded from the Canaries (T, G, H, P), in evergreen forest and pine forest mixed with *Erica-Myrica* community.

Phanerochaete sordida (P.Karst.) John Erikss. & Ryvarden *in* John Eriksson *et al., Cortic. North Europe* 5: 1023, 1978.
HABITAT AND DISTRIBUTION INSULAR.- **Tenerife (11)**, on decayed wood of *Euphorbia obtusifolia*; 5.XI.1994 (TFC Mic. 6941). **Tenerife (19)**, on decayed twigs of *Euphorbia obtusifolia*; 28.XII.1994 (TFC Mic. 7149).
REMARKS.- It is recorded from the Canaries (C, T, G, H, P), in evergreen forest, pine forest and pine forest mixed with *Erica-Myrica* community.

Phanerochaete tuberculata (P.Karst.) Parmasto, *Consp. Syst. Cortic.*: 83, 1968.
HABITAT AND DISTRIBUTION INSULAR.- **Tenerife (12)**, in humide *Rubus inermis-Salix canariensis* community, on fallen branches of *Salix canariensis* Chr. Sm. *ex* Link; 21.II.1992 (TFC Mic. 6392). **Tenerife (10)**, on decayed branches of *Euphorbia obtusifolia*; 14.XI.1994 (TFC Mic. 6885). **Tenerife (18)**, on unidentified dead wood; 12.XI.1994 (TFC Mic. 7138). **Tenerife (17)**, on decayed branches of *Euphorbia obtusifolia*; 12.XI.1994 (TFC Mic. 6945). **Tenerife (19)**, on the same substrate; 28.XII.1994 (TFC Mic. 7134).
REMARKS.- Previously reported for Hierro in *Myrica-Erica* forest.

Phanerochaete xerophila Burds., *Mycologia Memoir* 10: 141. 1985.
HABITAT AND DISTRIBUTION INSULAR.- **Tenerife (10)**, on decayed branches of *Euphorbia obtusifolia*; 14.XI.1994 (TFC Mic. 6886: Fig. 3).
REMARKS.- The Canary specimen is macroscopically similar to *P. tuberculata*, the margin is mainly fibrillose and unfringed. However its microscopic characters also resemble *P.xerofila*, described by Burdsall from xerophitic habitats of SW United States, where it seems to be common and frecuently confused with *P.tubercula* (Bursdsall, 1985); although differing from this by its subicular and subhymenial organization and wider subicular hyphae (x 8 µm.), without incrustations in the younger specimens. The spore dimensions are similar to those of *P.xerophila*, though the spores from the Canarian specimens are slightly wider and may reach 8 µm in diametre. *P.xerofila* has recently been reported from Portugal by Melo (1994), on decayed branches of *Erythrina caffra* Blanco.

Our specimen is also close to *Phanerochaete aurata* (H.Bourdot & Galzin) Burd., by its subicular and subhymenial organization. However it differs by its pubescent thin margin; the wider subicular hyphae (8 µm), ramified at right angles, occasionally encrusted; and the larger spore dimentions [-9 x (-6) µm].

Phlebiopsis ravenelii (Cooke) Hjortstam, *Windahlia* 17: 58, 1987.
HABITAT AND DISTRIBUTION INSULAR.- **Tenerife (13)**, on fallen leaves of *Phoenix canariensis*; 2.IV.1993 (TFC Mic. 6944).
REMARKS.- Recorded from the Canaries (C, T, G, H, P), in evergreen forest and pine forest

mixed with *Erica-Myrica* community.

Trechispora farinacea (Pers.: Fr.) Liberta, *Taxon* 15 (1): 318, 1966.
HABITAT AND DISTRIBUTION INSULAR.- **Tenerife (13)**, on fallen leaves of *Phoenix canariensis*; 2.IV.1993 (TFC Mic. 6956, 7135).
REMARKS.- Recorded in the Canaries (C, T, G, P), in evergreen forest and pine forest mixed with *Erica-Myrica* community.

Tubulicrinis gracillimus (Ellis & Ev. ex D.P. Rogers & H.S. Jackson) G. Cunn., *Bull. New Zeland Dept. Sci. Industr. Res.* 145: 141, 1963.
HABITAT AND DISTRIBUTION INSULAR.- **Tenerife (13)**, on fallen leaves of *Phoenix canariensis*; 2.IV.1993 (TFC Mic. 7137).
REMARKS.- Recorded in the Canaries (T, G, P), in evergreen forest, pine forest and pine forest mixed with *Erica-Myrica* community.

Ganodermataceae:
Ganoderma lucidum (M.A.Curtis: Fr.) P.Karst., *Rev. Mycol. (Paris)* 3 (9): 17, 1881.
HABITAT AND DISTRIBUTION INSULAR.- **Tenerife (12)**, in humide *Rubus inermis-Salix canariensis* community, on decayed wood of *Salix canariensis* Chr. Sm. *ex* Link; 21.II.1992 (TFC Mic. 6392).
REMARKS.- Recorded from Gran Canaria (*sine loc.*, Montagne, 1840), and in laurisilva of Tenerife.

Hymenochaetaceae:
Asterostroma cervicolor (Berk. & M.A.Curtis) Massee, *J. Linn. Soc. Bot.* 25: 155, 1890 [1889].
HABITAT AND DISTRIBUTION INSULAR.- **Lanzarote (1)**, on dead branches of *Nauplius intermedius*; 16.I.1994 (TFC Mic. 6921). **La Palma (22)**, on dead branches of *Euphorbia obtusifolia*; 8.XI.1991 (TFC Mic. 6400).
REMARKS.- Recorded for the laurel forests in Tenerife.

Inonotus tamaricis (Pat.) Maire, *Memor. Soc. Sci. Nat. Maroc.* 45: 84, 1937.
HABITAT AND DISTRIBUTION INSULAR.- **Fuerteventura (8)**, on living tree of *Tamarix canariensis*; 26.XII.1994 (TFC Mic. 6979). **Lanzarote (3)**: on fallen branches of the same substrate, along the road; 23.II.1993 (TFC Mic. 6931). **Gomera (20)**, on living tree of the same substrate; 4.III.1992 (TFC Mic. 6398).
REMARKS.- Known in the Canaries (L, C, T, P), on the same substrate.

Phellinus torulosus (Pers.) Bourdot & Galzin, *Bull. Soc. Mycol. France* 41: 191, 1925.
HABITAT AND DISTRIBUTION INSULAR.- **Tenerife (14)**, on unidentified decayed wood; 5.XI.1994 (TFC Mic. 6940).
REMARKS.- Known in the Canaries (T, G, P), in evergreen forest and pine forest mixed with *Myrica-Erica* community.

Polyporaceae:
Ceriporia purpurea (Fr.) Donk, *Konn. Nederl. Akad. Wet. Amst. Proc. Ser. C* 74 (1): 28, 1971.
HABITAT AND DISTRIBUTION INSULAR.- **Tenerife (11)**, on decayed wood of *Euphorbia obtusifolia*; 5.XI.1994 (TFC Mic. 6938). **La Palma (22)**, on dead branches of the same

substrate; 8.XI.1991 (TFC Mic. 6403).
REMARKS.- Recorded in the Canaries (C, T, P), in evergreen forest, pine forest and pine forest mixed with *Myrica-Erica* community.

Perenniporia medulla-panis (Jacq.: Fr.) Donk, *Persoonia* 5: 76, 1967.
HABITAT AND DISTRIBUTION INSULAR.- **Tenerife (12)**, on unidentified decayed wood; 2.IV.1993 (TFC Mic. 6599).
REMARKS.- Recorded in the Canaries (C, T), in evergreen forest.

Schizopora radula (Pers.: Fr.) Hallenb., *Mycotaxon* 18 (2): 308, 1983.
HABITAT AND DISTRIBUTION INSULAR.- **Tenerife (12)**, in humide *Rubus inermis-Salix canariensis* community, on decayed wood of *Salix canariensis* Chr. Sm. *ex* Link; 21.II.1992 (TFC Mic. 6392).
REMARKS.- Recorded from the Canaries (C, T, G, H, P), in evergreen forest and pine forest.

Schizophyllaceae:
Schizophyllum commune Fr., *Syst. Mycol.* 1: 330, 1821.
HABITAT AND DISTRIBUTION INSULAR.- **Tenerife (13)**, on fallen leaves of *Phoenix canariensis*; 2.IV.1993 (TFC Mic. 6602). **La Palma (22)**, on dead branches of *Euphorbia obtusifolia* and *Pinus canariensis*; 3.XI.1990, 28.VI.1991, 8.XI.1991, 3.IV.1992 (TFC Mic. 6181, 6388).
REMARKS.- Recorded by Jörstad (1966), in the semiarid-dry xerophytic areas of the infra-thermomediterrean bioclimatic belts: Barranco Hondo (Candelaria, T), on fallen branches of *Euphorbia obtusifolia*, and Barranco de Martinez (Orotava, T), on *Phoenix canariensis*. Also recorded by Cool (1924), from Güímar, on decayed cladodes of *Opuntia ficus-indica;* and by Bañares *et al.* (1980), in a *Rubia fruticosa-Juniperus turbinata* subsp. *canariensis* community (Sabinar de la Dehesa, H), in the dry thermomediterranean bioclimatic belt. This species has a wide altitude distribution, with its optimum in the forest areas of the humid-subhumid thermomediterranean and mesomediterranean bioclimatic belts (C, T, G, H, P).

DISCUSSION AND PROVISIONAL CONCLUSIONS

Analyzing the general ecology of the recorded species, only 3 may be classified as xero-thermophilic: *Phanerochaete xerofila, Inonotus tamaricis* and *Schizophyllum commune*, the latter with a wide altitude distribution; the remaining taxa have previously been recorded in the Islands forest ecosystems (evergreen and/or pine-forest). This is the case of *Phanerochaete andreae*, recently described from a mountain-top *Erica-Myrica* community of the thermomediterranean humid-subhumid belt (Parque Nacional de Garajonay, Gomera), widening its chorology and autoecology to the infra-thermomediterran arid-semiard bioclimatic belts of Lanzarote.

Collating our ecological data with other similar studies performed for the Iberian Peninsula, about Aphyllophorales with xero-thermophilic preferences [Honrubia & Llimona, 1979, 1982; Dueñas & Telleria, 1985; Ortega *et al.*, 1991, 1994; Vizoso *et al.*, 1991; auct. in Tellería (ed., 1992, 1993), etc.], we have confirmed that several of these Mediterranean species (16; 51.6%) are equally represented in these Canary habitats (*Byssomerulius corium, Globulicium hiemale, Hyphoderma praetermissum, H.sambuci, Peniophora boidinii,P.cinerea, P.incarnata, P.lycii, P.meridionalis, P.nuda, Phanerochaete martelliana, P.sordida, P.tuberculata, P.xerophila,Phlebiopsis ravenelii, Inonotus tamarici*). Nevertheless, Llimona *et al.* (1995), for a Catalonia area with similar climatic characterístics to ours, have collected Aphyllophorales, none of which are present in Canary arid-semiaride and dry habitats, although some of these species appear in more favourable environments in the Canary Islands. This may be attributed to the scattered collecting in theses habitats carried out up to now in the Canary Islands. Future explorations may probably record some most specific taxa like *Peniophora tamaricicola* Boidin & Malenç., on *Tamarix canariensis* Willd. and *Epithele canariensis* (Manjón & Moreno) Hjortstam, Manjón & Moreno, on *Phoenix canariensis* Chab., among others.

Euphorbia obtusifolia is the substract that supports the greatest number (11 species) of Aphyllophorales, followed by *Phoenix canariensis* (9 species).

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PHYLOGENETIC STUDIES IN GOMPHACEAE SENSU LATO (BASIDIOMYCETES)

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ABSTRACT

Phylogenetic relationships in Gomphaceae *sensu lato* were investigated. This has been a taxonomically difficult group of macroscopic fungi because of their heterogeneous external morphology, despite similarities in their microscopic features. Thirty-nine morphological characters were studied in nineteen species from ten genera. Five additional taxa represented the outgroups. A monophyletic group is circumscribed to include the genera *Gomphus, Gloeocantharellus, Kavinia, Beenakia, Ramaricium, Ramaria* and *Lentaria.* This monophyletic group is supported by a homoplasic synapomorphy (presence of mycelial cords). This clade was consistent under different combinations of outgroups and different character weightings and was assigned a family level. We propose the order Gomphales to include four families: Gomphaceae, Beenakiaceae, Ramariaceae and Lentariaceae. There is no resolution regarding which genus among potential outgroups is the sister group of the seven genera.

Key Words: phylogeny, Gomphaceae, taxonomy.

INTRODUCTION

The family Gomphaceae was established by Donk (1961) to include macroscopically heterogeneous fungi differing in their hymenial construction. In his classification the genera *Beenakia, Gomphus, Kavinia, Lentaria, Ramaria,* and *Ramaricium* in this family (see Table 1). Traditionally the Friesian system, classified these genera in the following four families: Cantharellaceae (pileate-stipitate basidioma and veins in the hymenium), Clavariaceae (coralloid basidioma with smooth hymenium and frequently amphigenous), Corticiaceae (resupinate basidioma and smooth to rough hymenia) and Hydnaceae (pileate-stipitate or resupinate basidioma with toothed hymenium). The classification by Donk (1961, 1964), contrary to previous ones, stressed microscopic similarities, for example, the presence of elliptic or elongated, cyanophilic spores, ochraceous or hyaline in color, and with a generally rugose wall. Another important characteristic of the Gomphaceae *sensu* Donk, is that the hymenium reacts positively with iron salts, producing a greenish color. Corner (1950) and Eriksson (1954) had already considered possible relationships between several pairs of taxa, for example *Gomphus* with *Ramaria; Lentaria byssieda* and *L. surculus* with species of the group of *Ramaria stricta,* and *Ramaricium* with *Ramaria* and *Kavinia*, due to the morphological similarity of their spores, their cyanophilic reaction or the form of the basidioma and the type of hyphal construction.

Several authors have recognized Gomphaceae *sensu* Donk (1961, 1964). Under this concept, genera like *Gloeocantharellus* and *Psathyrodon,* which previously had been considered in other groups, were also transferred to this family (Petersen, 1971a; Maas Geesteranus, 1971). However, the position of *Lentaria* within Gomphaceae is not clear, since it has been suggested that it should be excluded from Gomphaceae, but without indication as to where to place this genus (Estrada-Torres, 1994). Also, it has been recognized that genera from other families like *Clavariadelphus, Cantharellus,* and *Ramariopsis* show certain similarties with some of the members of Gomphaceae, but also with Clavariadelphaceae, Cantharellaceae or Clavariaceae. As a result, their affinities are considered intermediate among these four families within the Aphyllophorales (Corner 1950, 1966; Jülich, 1981; Methven, 1990; Petersen, 1971a, 1971b, 1988; Petersen *et al.,* 1974; Welden, 1966). On the other hand, there is also the question of whether to classify all of the genera within a single family or to classify them in many families in a single order. Thus, Julich (1981) created the order Gomphales, which was also recognized by Hawksworth *et al.* (1995). Nevertheless, as shown in table 1, discrepancies exist as to which families and genera are to be included in this group.

The generic and infrageneric clasification of Gomphaceae *sensu lato* has also been problematic. Currently, *Psathyrodon* is considered a synonym of *Beenakia*, (Parmasto and Ryvarden, 1990) and *Linderomyces* is a synonym of *Gloecantharellus*, (Petersen, 1971a).

Among the genera considered in the family or order Gomphales, *Ramaria* has the greatest number of species. More than one hundred species of *Ramaria* are currently known and it is estimated that *Ramaria* might include no less than two-hundred or three-hundred species (Petersen, 1971a; Marr & Stuntz, 1973; Estrada-Torres, 1994; Hawksworth *et al.*, 1995). *Ramaria* traditionally has been subdivided into three or four subgenera (*Lentoramaria, Ramaria, Echinoramaria,* and *Laeticolora*) which some authors consider to come from different ancestors, albeit all related to *Gomphus.* The other genera (*Beenakia, Ramaricium, Lentaria* and *Gomphus*) include less than twenty species and in some cases only one species (*Kavinia* and *Gloeocantharellus*).

Up until now there exists no taxonomical analysis with a rigorous phylogenetic approach that considers all these taxa. The objective of the present study was to evaluate alternative hypotheses for the classification of Gomphaceae *sensu lato* as a monophyletic group. Specifically, the genera *Gomphus, Ramaria, Kavinia, Beenakia, Lentaria, Ramaricium,,* and *Gloeocantharellus* were examined to determine if they constitute a natural group. The relationships of *Cantharellus, Clavariadelphus* and *Ramariopsis* were examined to determine if they also belong to Gomphaceae. As mentioned previously, different combinations of these genera have been ranked as families or orders, and at the same time, some genera have been included in other families or orders. For this reason, five taxa of diverse relationships (*Clavulina,* Auriculariaceae, Russulaceae, Amanitaceae and *Gymnopilus subpurpuratus*) were included as outgroups.

Genus	Donk (1961)	Julich (1981)				Hawksworth *et al.* (1995)			
	Aphyllophorales	Gomphales				Gomphales			Boletales
	A	A	B	C	D	A	B	C	D
Beenakia	●				●				●
Gomphus	●	●				●			
Kavinia	●				●			●	
Lentaria	●		●				●		
Ramaria	●			●				●	
Ramaricium	●				●			●	
Gleocantharellus		●				●			
Linderomyces		●							
Delentaria			●					●	
Psathyrodon					●				
Pseudogomphus						●			
Terenodon						●			

Families: A = Gomphaceae; B = Lentariaceae; C = Ramariaceae; D = Beenakiaceae

Table 1. Traditional classifications of Gomphaceae *sensu lato* as proposed by different authors.

MATERIALS AND METHODS

Phylogenetic analyses. Information derived from the present morphological survey was interpreted and analysed by means of cladistic methods (Forey *et al.,* 1992). These analyses start with the procedures for character selection, state recognition, and codification on a data matrix. Morphological characters are evaluated first with the empirical criteria of similarity, discrete variation, conjunction, heritability and independence (De Pinna, 1991; De Luna and Mishler, 1997). Characters were also subjected to the congruence tests using the inferential criterion of parsimony. After a phase of character analyses follows the search for cladograms (Kitching, 1992). These were performed with PAUP (Swofford, 1991). Given the 24 units of comparison, exhaustive searches are not possible. The only available option was to execute heuristic searches under different strategies as suggested by Soltis and Soltis (1996). The interpretation of a cladogram as a phylogenetic hypothesis is only possible until levels of clade support have been examined. A phylogenetic hypothesis of the Gomphales was used to propose a classification of families within this order.

Characters were obtained from the study of herbarium specimens and with the support of data gathered from several bibliographical sources (Cifuentes 1996; Corner 1950, 1970, 1966; Estrada-Torres 1994; Heim 1959; Keller 1986; Marr & Stuntz 1973; Maas Geesteranus 1963, 1967, 1971, and 1977; Methven 1990; Parmasto & Ryvarden 1990; Pegler & Young 1985; Petersen 1967a, b, 1968, 1969, 1971a, 1971b, 1971c, 1972, 1974, 1975, 1976, 1978, 1985, 1987, 1988; Reijnders & Stalpers 1992; Singer 1945; Sung 1987; Welden 1966). Representative samples of the six genera considered by Donk 1961 and 1964) in the family Gomphaceae were revised. *Gloeocantharellus* was also included, which was later considered within Gomphaceae *sensu lato* by authors like Petersen (1971a), Julich (1981), and Hawksworth *et al.* (1995) (see Table 1). Three genera (*Clavariadelphus, Ramariopsis* and *Cantharellus*) that may be related to this family as considered by other authors were also revised (Corner 1966, 1970; Jülich 1981; Methven 1990; Petersen 1967, 1971b, 1973, and 1988; Welden 1966). In total, 146 samples were studied from ten genera. These specimens are deposited in the herbaria at: Facultad de Ciencias, UNAM, Mexico (FCME), University of

Tennessee, Knoxville, EUA (TENN), Centro de Investigacion en Ciencias Biologicas, Universidad Autónoma de Tlaxcala, Mexico (TLXM), Escuela Nacional de Ciencias Biologicas, Instituto Politécnico Nacional, Mexico (ENCB), Instituto de Botánica, Universidad de Guadalajara, Mexico (IBUG), Instituto de Biología, UNAM, Mexico (MEXU), and Instituto de Ecologia, A.C., Xalapa, Mexico (XAL). The ten genera considered for this study were represented by nineteen species to sample infrageneric variation. It has been problematic to obtain representatives of genera like *Ramaricium,* where only one sample from India was revised and where support was found in the scientific literature (Jülich, 1979; Sung, 1987; Ginns, 1969). Other genera considered in Table 1 like *Terenodon, Pseudogomphus* and *Delentaria* were not included in the present analysis for various reasons. Only the type specimen of *Terenodon* is known and it was not available for examination. The record in Mexico like *Terrenodon serenus,* (Guzman 1983) has not been verified (Cifuentes 1996). Specimens of *Psuedogomphus* and *Delentaria* have not been found.

Outgroups. The present cladistic analysis was designed to explore the monophyly of Gomphaceae *sensu lato*, which traditionally have included seven genera: *Beenakia, Gomphus, Kavinia, Lentaria, Ramaria, Ramaricium* and *Gloeocantharellus.* We also tested the inclusion in this family of three additional genera: *Cantharellus* (Cantharellaceae), *Clavariadelphus* (Clavariadelphaceae), and *Ramariopsis* (Clavariaceae). As a potential outgroup we included *Clavulina* (Clavulinaceae). The relations of these families have not been evaluated cladistically, so that the sister group of Gomphaceae *sensu lato* is unknown. In order to properly orient trees including the above collection of ten genera, a set of distant outgroups was also necessary. Four additional Operational Taxonomic Units (OTUs) were considered to represent the variation of the Agaricales, Russulales, and Auriculariales. The Agaricales was represented by two OTUs, one was reconstructed with the states present in the Amanitaceae and the second with those of the genus *Gymnopilus* (Cortinariaceae). In a similar fashion, the Russulales was represented using the known states in the Russulaceae and the Auriculariaceae was sampled to study the Auriculariales.Various characters were found to be polymorphic, for example, in both Russulaceae and Amanitaceae, the reaction to ferrous sulphate is positive in some species and negative in others. An initial global analysis with all taxa and all outgroups included strongly suggested the exclusion of *Clavariadelphus, Cantharellus,* and *Ramariopsis* from Gomphaceae *sensu lato*. These taxa were thus considered as additional outgroups in further analyses to test the effect of different combinations of close and distant outgroups.

Character Analysis. The selection of characters was based on the observed variation among the different genera and guided by a phylogenetic concept of homology (De Pinna, 1991). Previous taxonomical proposals of this group were reviewed in order to obtain potentially useful characters. Nevertheless, many of these features are not found with a clear and homogeneous description at the generic level. Thus, many of the features used in the present analysis were obtained through the observation of herbarium specimens or, in some cases, with fresh samples.

The selection of potentially useful characters was based on the evaluation and explicit examination of each one of them. In terms of the levels of variation within and among OTUs. Some characters like color, basidioma context, smell, taste, and basal mycelium were not included in the present analysis due to incomplete information or to the fact that character variation is not discrete. In some OTUs like *Gomphus floccosus, Gomphus clavatus, Gloeocantharellus purpurascens, Cantharellus cibarius,* and *Clavariadelphus truncatus*, ontogenetic data of the hymenial surface were available, but were excluded from the analysis since they were not known for all OTUs. For this reason, only the mature state of the basidioma was coded. Nevertheless, these data confirm the association of different states observed as part of the same character state system. There are other characters in which the variation could not be partitioned into character states like the coloration of the spore mass, their form and ornamentation, and changes of color when manipulated or exposed.

A total of thirty-nine informative characters were considered, which include the macro- and microscopic morphology of the different basidia that were examined (Table 2). The microscopic survey was done using standard techniques. A study of the spores using a scanning electron microscope is in process. All multistate characters are unordered, since no information was available to establish clearly the relationship between states.

Cladistic Analyses. Four sets of cladistic analyses were executed with PAUP 3.0 (Swofford 1991). The data matrix (Table 3) consisted of twenty-four taxa and thirty-nine characters in which

Characters	Character States
1. Basidioma pileate	0 = present, 1 = absent
2. Basidioma effused	0 = present, 1 = absent
3. Basidioma clavate	0 = present, 1 = absent
4. Basidioma branched	0 = simple, 1 = little branched (1to 3 levels of ramification), 2 = very branched (more 3 levels of ramification), ? = ambiguous
5. Hymenophore with teeth	0 = present, 1 = absent
6. Hymenophore with gill-folds or ridged	0 = absent, 1 = not very deep and reticulate, 2 = deep
7. Hymenophore without projections	0 = smooth, 1 = rugose (at least partially), ? = does not apply
8. Hymenophore with lamellae	0 = present, 1 = absent
9. Stipe	0 = clearly differentiated, 1 = little differentiated, 2 = absent
10. Stipe position regarding to the cap	0 = central, 1 = eccentric, ? = does not apply
11. Basidioma always gelatinous	0 = present, 1 = absent
12. Basidioma changing color on bruising or cutting	0 = present, 1 = absent
13. Macrochemical reaction to iron salts on fresh basidioma	0 = present, 1 = absent
14. Macrochemical reaction to iron salts on dried basidioma	0 = present, 1 = absent
15. Conspicuous mycelial cords	0 = present, 1 = absent
16. Subiculum	0 = present, 1 = absent
17. Types of basidia	0 = holobasidia, 1 = phragmobasidia
18. Basidia transversely septate at maturity	0 = present, 1 = absent
19. Number of sterigmata per basidium	0 = two, 1 = two or more
20. Type of basidia regarding the orientation of achromatic spindle	0 = chiastic, 1 = stichic, ? = unknown
21. Color of spores under a microscope	0 = hyaline, 1 = pale cream to ochraceous
22. Spore shape	0 = globose, 1 = non globose
23. Spore ornamentation	0 = smooth, 1 = rugose, 2 = echinulate
24. Spore wall surface	0 = irregular, 1 = striate
25. Cyanophilous ornamentation in spores	0 = present, 1 = absent
26. Spores amyloid	0 = present, 1 = absent
27. Spores dextrinoid	0 = present, 1 = absent
28. Clamp connections uninflated in the basidioma	0 = present, 1 = absent
29. Inflated clamp connections in the basidioma	0 = present, 1 = absent, ? = unknown
30. Inflated clamp connections delicately ornamented	0 = present, 1 = absent, ? = does not apply
31. Basidia with clamps	0 = present, 1 = absent
32. Position of hypha in subhymenium	0 = interwoven, 1 = parallel
33. Coscinoidal hyphae	0 = present, 1 = absent
34. Gloeoplerous hyphae	0 = present, 1 = absent
35. Cystidia	0 = present, 1 = absent
36. Cystidia wall thickness	0 = thin, 1 = thick, ? = does not apply
37. Cystidia content	0 = homogeneous appearence, 1= with vacuolized or oily contens, ? = does not apply
38. Position of cystidia regarding basidia	0 = intercalate, 1 = not intercalate, ? = does not apply
39. Size of cystidia regarding basidia	0 = same or smaller size, 1 = projecting out conspicuously

Table 2. Morphological characters and character states used for cladistic analyses of Gomphaceae *sensu lato*.

	Character and character States
	111111111122222222223333333333
Taxa	123456789012345678901234567890123456789
Ramaria grandis	11121001101000110100112001100100100????
	1 1
Ramaria gracilis	11121001101100010110111001100000110????
Ramaria cf. *botrytis*	11121001101000110110111101100?00100????
Gomphus floccosus	011011?100100011011011100111001000000000
	1
Gomphus clavatus	111111?10010001101101110011100000100????
Kavinia alboviridis	101000?12?1?00000110111001110101110????
	1
Beenakia fricta	011000?1011000000110111001110101110????
Lentaria surculus	1112100110110000011001 0?1111010?110????
	1
Gloeocantharellus purpurascens	011012?100100011011011100111 11?000010100
Clavariadelphus truncatus	11001011101000110110010?111101011010???
Cantharellus cibarius	011012?1001000110111010?11111?00110????
	1
Ramariopsis kunsei	11121001101111110110002011111?00110????
Ramariopsis corniculata	111?1001101100110110000?11111?00100????
	1
Ramaria abietina	11121001101000010110112001110100100????
	1
Ramaria stricta	11121001101000010110111001110100110????
Ramaricium sp.	101010012?1?0?11011010200111010 1110????
Ramaria cf. *fennica*	11121001101000110110111001100100110????
	1
Ramaria rubripermanens	11121001101000110110111101100?00110????
Clavariadelphus pistillaris	11001001101100110110010?111001001110???
Gymnopilus subpurpuratus	011011?00010111101 1?1110?10011?01110010
Russulaceae	011010?000100?000110002010101?10??10000
	11 11 1 1 1111
Auriculariaceae	011010012?011111111?010?11101?001110000
	1
Amanitaceae	011010?0001001010100000?10101?000001000
	1 1 1 1 1 1 1 111 1
Clavulina	10101001101?11110000000?11101?001000001
	1 1 1 1 11 11 1
	2

Table 3. Datamatrix based on 39 morphological characters for 19 taxa of Gomphaceae *sensu lato*

the autapomorphies were also considered. Tree search was always done with autapomorphies removed. Tree lengths are reported both with and without autapomorphies. It was only possible to perform heuristic searches in the majority of analyses. This type of search typically explores only subgroups of all the possible trees. Nevertheless, to avoid exploring only one island, multiple searches with random additions of taxa (ten replicates) were made in combination with the TBR swapping algorithm in PAUP. Four series of analyses were developed to find trees under different exploration conditions as described in table 4.

In all cases, the topologies were directed with one of the outgroups, which were included according to each of the tree search conditions (Table 4). A strict consensus of the trees was constructed to show monophyletic groups present in all of the equally most parsimonious trees found in each of these five series of analyses. Optimization of character states was made utilizing the ACCTRAN option in PAUP. The final results and interpretations on the number of character changes in each branch were examined on one of the most parsimonious trees found in the first series of analysis (Figure 1). To evaluate the relative robustness of groups within the trees, decay analyses were made as described in Bremer (1988). Trees of up to three steps longer than the most parsimonious ones were saved to estimate the decay index of the different clades of each one of the analyses. These indices were calculated on strict and 80 % majority consensus.

RESULTS

Phylogenetic relationships. The topologies found in the first series of analyses (using the TBR algorithm for branch exchange) are summarized in the strict consensus tree shown in Figure 2a. A total of six equally parsimonious trees of 105 steps (131 with autapomorphies) were found, with a consistency index (CI) = 0.657, a retention index (RI) = 0.670, and a rescaled index (RC) = 0.440. In these analyses, seven genera (*Ramaria, Lentaria, Beenakia, Ramaricium, Kavinia, Gomphus,* and *Gloeocantharellus*), are found to make up a monophyletic group. Nevertheless, there is no resolution regarding the sister group of this clade, since it can be the clade formed by *Ramariopsis* and *Clavulina* or *Clavariadelphus*. Within Gomphales *sensu lato*, a relevant clade is the one that includes *Gomphus* and *Gloeocantharellus.*

In the second series of analyses, all of the outgroups were eliminated (represented by ten OTUs) and then only species of seven genera of Gomphaceae were considered. With this reduced matrix, it was possible not only to perform the heuristic search, but another search with the branch and bound algorithm. Both searches were performed giving the same weight to all characters, and as a result, we found one tree of thirty-eight steps (without autapomorphies, CI = 0.737, RI= 0.722, and RC = 0.532). Taking into account the results of the previous analysis 1, the tree was oriented with *Lentaria.* Relationships among genera in this tree are identical to those in the clade of Gomphaceae in analysis No. 1 (Figure 2b).

In analyses numbers three and four, the four farthest or nearest outgroups were eliminated alternatively (Table 4). The heuristic search with the nearest outgroups (analysis four) with twenty OTUs resulted in two trees (Table 5). The consensus shows two resolved monophyletic groups, which includes the seven genera of Gomphaceae *sensu lato.* There is no resolution as to which genus is the sister group to this clade. A search with branch and bound algorithm was also performed, finding the same two trees. The heuristic search (analysis 4) with eighteen OTUs and a search with branch and bound revealed one hundred and four more parsimonious trees. The same monophyletic group of seven genera stands out once again in the strict consensus tree. Just as in analysis number three, the relationships between the outgroups and Gomphaceae is ambiguous (Figure 3b).

Robustness analysis. The resulting trees of each one of the four cladistic analyses were evaluated by the decay index (Bremer 1988). The estimated indices of branch support in the five analyses fluctuate between one and two. The majority of the clades are collapsed in the strict consensus trees that are only one step longer than the most parsimonious one. The most robust clade is the one that includes the seven genera of Gomphaceae, although the subgroup that excludes *Lentaria surculus* is also robust. Within Gomphaceae the clades which incorporate *Gomphus floccosus* and *Gloeocantharellus purpurascens* and the one which includes *Kavinia albovirdis* and *Beenakia fricta*, are also robust.

Analysis	**OTU's**	**Search**	**Swapping algorithm**
1	Ingroup and both outgroups	Heuristic	TBR, SPR and NNI
2	Only the ingroup and *Lentaria* as outgroup	Heuristic and "Branch & Bound"	TBR, SPR and NNI
3	Ingroup and closest outgroup	Heuristic and "Branch & Bound"	TBR, SPR and NNI
4	Ingroup and farthest outgroup	Heuristic and "Branch & Bound"	TBR, SPR and NNI

Table 4. Conditions of exploration used in the different analyses.

Analysis	**Search**	**Trees obtained**	**Length of most parsimonious tree**	**Indices**
1a	Heuristic with SPR and TBR	6	105	CI = 0.657 RI = 0.670 RC = 0.440
1b	Heuristic with NNI	562	130	CI = 0.692 RI = 0.633 RC = 0.438
2	Heuristic with TBR, SPR and NNI Branch & Bound	1	38	CI = 0.737 RI = 0.722 RC = 0.532
3	Huristic with TBR, SPR and NNI Branch & Bound	2	73	CI = 0.658 RI = 0.679 RC = 0.447
4	Heuristic with TBR,SPR and NNI Branch &Bound	104	99	CI = 0.768 RI = 0.647 RC = 0.504

CI= Consistency Index, RI= Retention Index, RC= Rescaled consistency Index.

Table 5. Results obtained with different search options. In duplicated analyses (1a,1b) the data matrix content (OTU's and characters) is identical. The data matrix is not same among analysis due to different combinations of outgroups. In all analyses, heuristic search were performed with random addition of taxa and 10 replicates.

DISCUSSION

The results obtained in the four analyses (Figures 2-3) suggest that the seven genera *Gomphus, Gloecantharellus, Kavinia, Beenakia, Ramaricium, Ramaria,* and *Lentaria* form a robust monophyletic group. The final results and interpretations are illustrated in one of the trees found in analysis one (Figure 1). This cladogram identifies the monophyletic group of seven genera which is recovered under four distinct conditions of analyses in which the effects of various combinations of outgroups were evaluated. From these four analyses, it can be also noticed on the strict consensus trees that *Cantharellus, Clavariadelphus,* and *Ramariopsis* do not belong to this monophyletic group under any of the evaluated circumstances. This result is relevant since the inclusion of these genera had been suggested by different authors (Corner, 1966; Jülich, 1981; Methven, 1990; Petersen, 1971a, 1971b and 1988; Petersen *et al.*, 1974; Welden, 1966; Marr & Stuntz 1973). The examination of individual trees from analysis one shows that *Clavariadelphus*-plus- Gomphales form a monophyletic group (Fig. 1). However, this sister group relationship is not present in all trees (Fig. 2a).

On the other hand, although these analyses were not designed to elucidate the relationships among the seven genera within the clade Gomphaceae *sensu lato*, the topology allows us to explore the following groups (Fig.1). One group includes *Gomphus* and *Gloeocantharellus.* The second group consists of *Kavinia, Beenakia,* and *Ramaricium.* The third one corresponds to the species of *Ramaria*, which although it is not monophyletic in any of the present analyses, the possibility of its existence as a group is suggested by an additional analysis, in which we included all of the OTUs, (heuristic search) and retained a set of subparsimonious trees (one step longer than the shortest one). Some of these trees recover all of the species of *Ramaria* as a monophyletic group.

The Gomphales (Fig. 1) is suppported by only one synapomorphy: the presence of evident mycelial cords (15:0). Within this same clade, another three synapomorphies allow us to recognize one subgroup of six genera, out of which *Lentaria* is excluded. One (character 25, spores with cyanophylic ornamentation) is shown as a unique synapomorphy and the other two (21, spores in mass with brown coloration and 23 spores with ornamented wall) are not unique, but are interpreted as independently derived for Gomphales. The results of this study strongly support the hypothesis proposed by Jülich (1981) and partially by Donk (1964). This was also followed by Estrada-Torres (1994), but including *Gloecantharellus* and excluding *Lentaria*, since the last two authors only considered Gomphaceae as a single family (Table 1). Their interpretations are based on characteristics like the reaction of the hymenium to iron salts, the cyanophillic character of spores, and their coloration and ornamentation.

With the results obtained in this analysis, it is not possible to make inferences regarding relationships within Gomphales, since this requires other methodological considerations. However, in genera like *Ramaria* where seven representative samples were included, all the analyses revealed this group as polyphyletic. *Ramaria*, with its branched basidioma and brown color and the ornamented spores, traditionally has been considered as one single genus, but subdivided into four subgenera (*Ramaria, Laeticolora, Lentoramaria, and Echinoramaria*). However, some authors have suggested that each subgenus could have originated from different ancestors, corresponding thus to different genera (Petersen 1967a, 1970, 1971a, and 1981; Marr and Stuntz 1973). The present series of analyses reveals that the subgenus *Ramaria* (*Euramaria*), represented here by *R.* group *botrytis* and *R. rubripermanens*, is recovered as a monophyletic group by the majority of analyses, which suggests a natural support for the taxonomical recognition of the subgenus *Ramaria (Euramaria).* The same analyses convey other subgenera as paraphyletic. Futhermore they show no close relationship with members of the genus *Gomphus* as has been suggested by Petersen (1967b, 1970, 1971a). Further research should include additional species and characters for detailed cladistic analyses within *Ramaria* as presently defined. Meanwhile, even though the seven representatives of *Ramaria* do not form a monophyletic group, it is still convenient to classify them in the Ramariaceae (Fig. 1). Traditionally, Gomphaceae *sensu lato* has been taxonomically recongnized at two levels: as a family (Gomphaceae) and as an order (Gomphales). The phylogenetic analyses presented here (Fig. 1), support the placement of the seven genera present in a monophyletic group. We propose the recognition of this clade at the ordinal level as Gomphales. This clade in turn can be subdivided into four different groups: the first one made up by the genera *Gomphus* and *Gloeocantharellus*, the second by *Kavinia, Beenakia* and *Ramaricium*, the third by the genus *Ramaria* and the fourth by *Lentaria*. We propose the recognition of these four clades at the family level to recognise the Gomphaceae, Beenakiaceae, Ramariaceae and Lentariaceae. The inclusion of *Clavariadelphus* within Gomphales is not in conflict with monophyletic groups revealed by present analyses (Fig. 1). Based on this alone, it would be only a difference of ranking. However, the clade that includes *Clavariadelphus* plus the Gomphales is not stable in neither of the strict consensus of several analyses performed here (Figs. 2a and 3a). Furthermore this clade is not robust as suggested by decay analyses (Fig. 2a). With molecular dates Hibbett (1977) obtained similar results but he suggest that *Clavariadelphus* is closely related to "gomohoid lineage".

Hawksworth *et al.*, (1995) have considered that the genus *Beenakia* can be placed in a different order (Boletales), the justification for this proposal is not stated. A review of the relevant literature on the Beenakiaceae and Boletales leads to the conclusion that *Beenakia* was not classified in the Boletales but until Hawksworth *et al.* (1995). Indeed Jülich, (1981, 1983) have not even the posibility of such a transfer. Neither Bruns *et al.* (1990) or Besl & Bresinsky (1997), in their studies of the Boletales regarded *Beenakia* as a member of the order. Furthermore the present cladistic analysis suggest that the inclusion of *Beenakia* in the Boletales seems unwarranted at present. It is still possible that spore morphology might be similar between *Beenakia* and some families in the Boletales. However, to evaluate the phylogenetic relationships of *Beenakia* further analyses should include genera like *Boletellus, Boletus* and other Boletales. The cladistic analyses reported here suggest a relationship of *Beenakia* with *Kavinia,* and of these two with *Ramaricium*. Nevertheless, our results cannot establish whether this suggested relationships is an artifact due tu sampling of taxa or a real phylogenetic pattern. This situation illustrates the need to use other outgroups in the future.

AKNOWLEDGEMENTS

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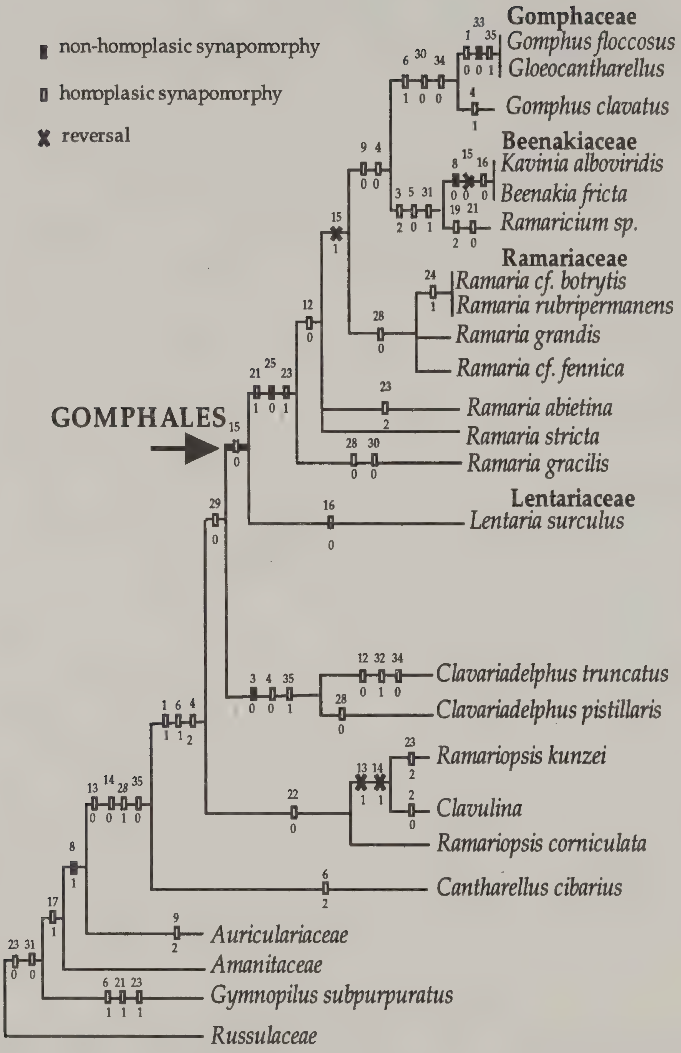
▮ non-homoplasic synapomorphy
▯ homoplasic synapomorphy
✖ reversal
Gomphaceae
Gomphus floccosus
Gloeocantharellus
Gomphus clavatus
Beenakiaceae
Kavinia alboviridis
Beenakia fricta
Ramaricium sp.
Ramariaceae
Ramaria cf. botrytis
Ramaria rubripermanens
Ramaria grandis
Ramaria cf. fennica
Ramaria abietina
Ramaria stricta
Ramaria gracilis
GOMPHALES
Lentariaceae
Lentaria surculus
Clavariadelphus truncatus
Clavariadelphus pistillaris
Ramariopsis kunzei
Clavulina
Ramariopsis corniculata
Cantharellus cibarius
Auriculariaceae
Amanitaceae
Gymnopilus subpurpuratus
Russulaceae

Fig.1. This is one tree selected at random from the set of six most parsimonious trees obtained in the analyses one. This tree shows the basic clades and character changes.

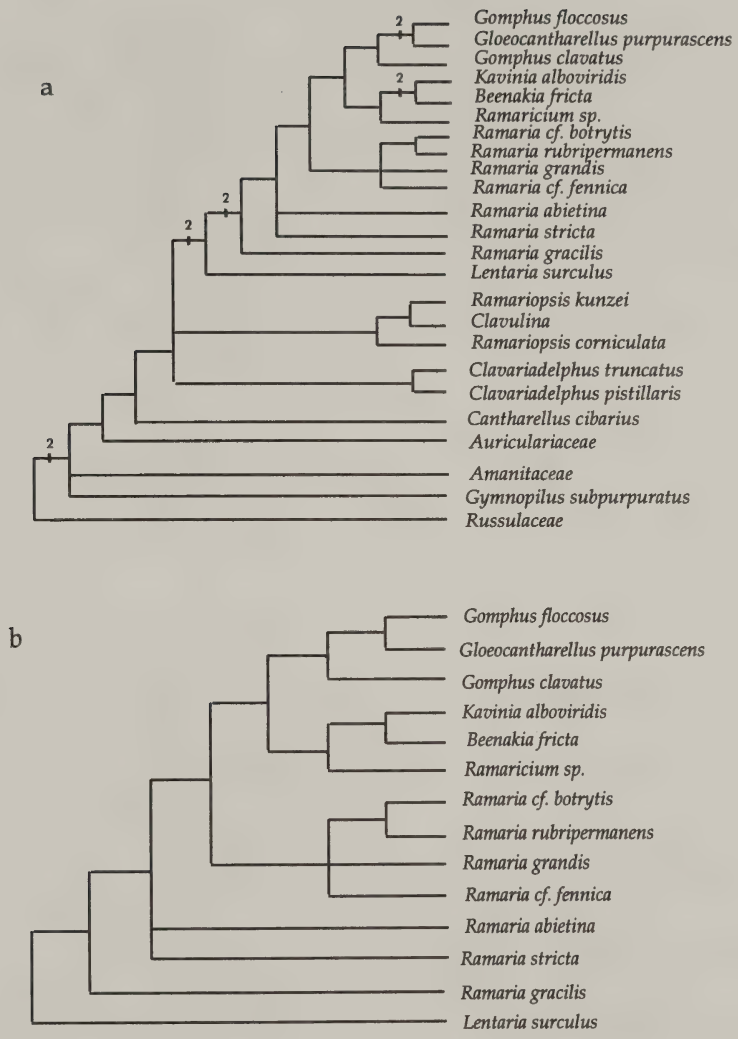
a
2
2
2
2
2
Gomphus floccosus
Gloeocantharellus purpurascens
Gomphus clavatus
Kavinia alboviridis
Beenakia fricta
Ramaricium sp.
Ramaria cf. botrytis
Ramaria rubripermanens
Ramaria grandis
Ramaria cf. fennica
Ramaria abietina
Ramaria stricta
Ramaria gracilis
Lentaria surculus
Ramariopsis kunzei
Clavulina
Ramariopsis corniculata
Clavariadelphus truncatus
Clavariadelphus pistillaris
Cantharellus cibarius
Auriculariaceae
Amanitaceae
Gymnopilus subpurpuratus
Russulaceae
b
Gomphus floccosus
Gloeocantharellus purpurascens
Gomphus clavatus
Kavinia alboviridis
Beenakia fricta
Ramaricium sp.
Ramaria cf. botrytis
Ramaria rubripermanens
Ramaria grandis
Ramaria cf. fennica
Ramaria abietina
Ramaria stricta
Ramaria gracilis
Lentaria surculus

Fig. 2. a) Strict consensus obtained in the analyses 1 and b) tree obtained in the analyses 2. Estimated values of the "decay index" are given on the branches (a)

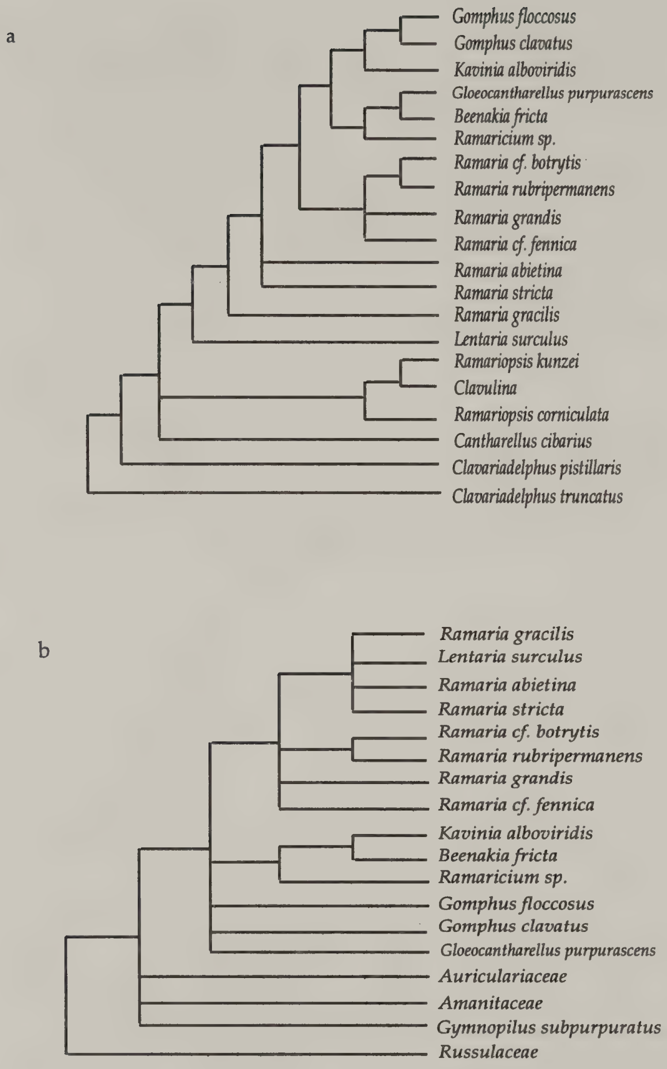
a
Gomphus floccosus
Gomphus clavatus
Kavinia alboviridis
Gloeocantharellus purpurascens
Beenakia fricta
Ramaricium sp.
Ramaria cf. botrytis
Ramaria rubripermanens
Ramaria grandis
Ramaria cf. fennica
Ramaria abietina
Ramaria stricta
Ramaria gracilis
Lentaria surculus
Ramariopsis kunzei
Clavulina
Ramariopsis corniculata
Cantharellus cibarius
Clavariadelphus pistillaris
Clavariadelphus truncatus
b
Ramaria gracilis
Lentaria surculus
Ramaria abietina
Ramaria stricta
Ramaria cf. botrytis
Ramaria rubripermanens
Ramaria grandis
Ramaria cf. fennica
Kavinia alboviridis
Beenakia fricta
Ramaricium sp.
Gomphus floccosus
Gomphus clavatus
Gloeocantharellus purpurascens
Auriculariaceae
Amanitaceae
Gymnopilus subpurpuratus
Russulaceae

Fig. 3. a) Strict consensus obtained in the analyses 3. b) Strict consensus obtained in the analyses 4. (See table 5 for details)

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NATS TRUFFLE AND TRUFFLE-LIKE FUNGI 8: RHIZOPOGON MENGEI SP. NOV. (BOLETACEAE, BASIDIOMYCOTA)

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ABSTRACT

A new sequestrate fungal species associated with Adenostoma fasciculatum is described as Rhizopogon mengei. It differs from the related species, R. ventricisporus, in spore size and shape, lack of peridial staining where bruised and lack of peridial intracellular and encrusting pigment in KOH mounts. This is the first confirmed report of a Rhizopogon sp. associated with an angiosperm in nature.

INTRODUCTION

Studies of the ecology and mycorrhizae of chamise (Adenostoma fasciculatum Hook. & Arn.) in southern California in collaboration with Edith B. Allen led to the discovery of a hypogeous fungus that appeared macroscopically to be a Rhizopogon. Lack of Pinaceae as suitable mycorrhizal hosts for Rhizopogon spp. at the collection site suggested the fungus might belong to a different genus. Microscopic examination revealed it to have typical Rhizopogon features, however, and DNA analysis confirmed it to be a Rhizopogon. Accordingly, we describe it as a new species for that genus.

METHODS

Colors of fresh specimens are in general terms. After notes were taken on fresh specimens, they were dried on a forced air dehydrator set at 49°C (120°F). Microscopic characters were determined from hand sections mounted in 5% KOH or Melzer's reagent as indicated. The holotype has been accessioned into the Mycological Herbarium of Oregon State University (OSC). Isotypes and paratypes have been accessioned into the H. D. Thiers Herbarium of San Francisco State University (SFSU).

TAXONOMY

Rhizopogon mengei M. Allen, Trappe & Horton, sp. nov. Fig. 1.

Basidiomata 6-30 x 10-45 mm., subglobosa, lutea vel brunnea, rhizomorphis adpressis, albis vel concoloratis. Gleba cartilaginea, pallide avellanea, loculis 0.1-0.4 mm latis, rotundis vel oblongis et sporis repletis; columella absens. Peridium usque ad 200 µm crassum, hyphis hyalinis vel pallide luteis, 3-5 µm in diam, cellulis dispersis inflatis usque ad 4-6 (-12) µm; pigmentum absens. Sporae 7-12 (-15) x (3.5-) 4-6 .5 µm, ellipsoideae vel oblongae, laeves, hyalinae. Adenostomate consociatus. Holotypus Trappe 17946.

Basidiomata 6-30 x 10-45 mm, subglobose, yellow to brown, not staining where bruised, with scattered, appressed, white to concolorous rhizomorphs. **Gleba** rubbery, white in youth, becoming light yellowish brown, the color a combination of white to brown trama and light brownish yellow spore masses filling the rounded to oblong locules 0.1-0.4 mm broad; columella lacking. Chemical reactions: 5% KOH on peridium brown, 5% $FeSO_4$ faintly olive.

Odor and **taste** not distinctive.

Peridium 75-100 µm thick in young specimens, up to 200 µm thick by maturity, of appressed-interwoven, hyaline to pale yellow, thin-walled hyphae 3-5 µm in diam (the "*roseolus*-type" of Martin, 1996), scattered cells inflated to 4-6 (-12) µm, clean (lacking pigment deposits) in KOH; Melzer's reagent nonreactive; as dried lacking any red tones. Clamp connections not found.

Glebal trama with a center of subparallel, hyaline hyphae 3-6 µm in diam with gelatinous-thickened, glassy-appearing walls at maturity, the zones between the central tissue and subhymenium of similar but interwoven hyphae that diverge to the **subhymenium** of ± isodiametric cells 5-9 µm in diam. **Basidia** thin-walled, clavate, 12-15 (-24) x 5-8 µm, with interspersed ellipsoid, hyaline, thin-walled brachybasidioles 15-20 x 7-12 µm; sterigmata not seen.

Spores 7-12 (-15) x (3.5-) 4-6.5 µm, ellipsoid to oblong (rare spores in some specimens are narrowly obovoid or peanut-shaped), smooth, thin-walled, the basidial attachment indistinct and 1.5-2 µm broad, in youth with 1-2 large and several small guttules, in age eguttulate, in KOH hyaline singly and in mass brownish yellow, in Melzer's reagent light yellow singly and in mass brownish yellow, in cotton blue light to moderate blue.

Etymology: In honor of Dr. John Menge, who has contributed much to knowledge of the southern California mycoflora and was convinced of the need to further explore the mycorrhizal fungi of arid-land shrubs.

Collections examined: HOLOTYPE—**California,** Riverside Co.: Lake Skinner, M. Allen #131, Trappe #17946, March 1996 (OSC 59792, Isotype SFSU). PARATYPES-**California,** Marin Co.: Mt. Tamalpais, Rock Springs Area, Simmon's Trail, Tom Horton #200, 21 Jan. 1992 (OSC 59793), #201, 9 Dec. 1992 (OSC 59794), #202, 9 Jan. 1993 (OSC 59795). Riverside Co.: Lake Skinner (type locality), M. Allen, Trappe #17823, 19 Feb. 1996 (OSC 59791, SFSU). San Diego Co..: Anza Borrego State Park, Sky Oaks Biological Field Station, Coumbs Camp, A. Jumpponen, Trappe #13287, 20 March 1994 (OSC 59790).

Distribution, habitat and season: hypogeous, California (Riverside and Marin Counties) The type locality is a pure stand of *Adenostoma fasciculatum* Hook. & Arn. at elev. ca. 420 m with no Pinaceae or Fagaceae within a km. The other collections were in *Adenostoma* stands in mixture with scattered other woody plants *(e.g., Arctostaphylos montana, Quercus dumosa, Pinus coulteri).*

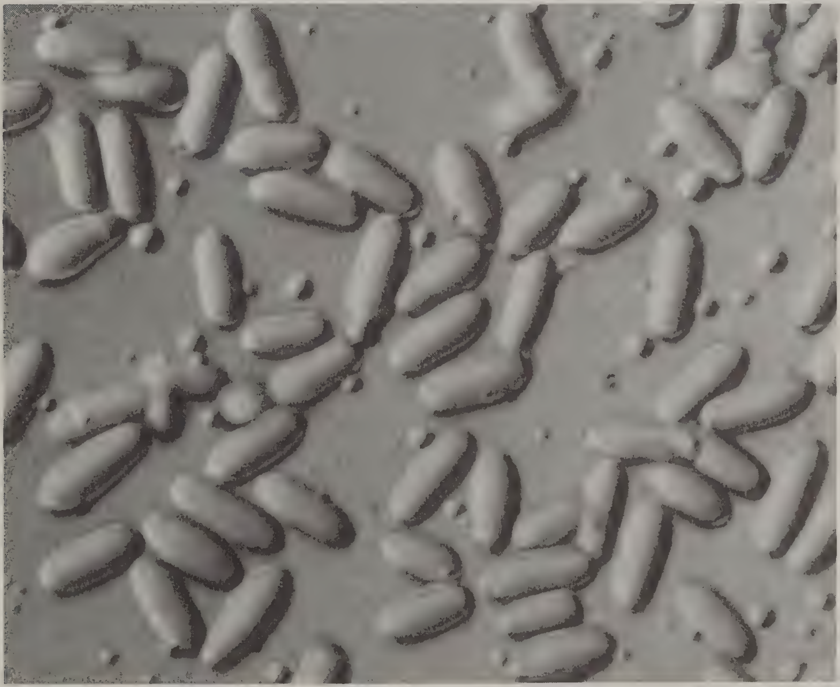

Fig. 1. Spores of *Rhizopogon mengei* mounted in 5% KOH., x 1,500. Note large variation in spore length.

Discussion: Even relatively young specimens had so many spores massed on the hymenial surface that it was impossible to determine how many spores are borne on individual basidia. If sterigmata were present on basidia, they were too small to detect by light microscopy, as is the case with most *Rhizopogon* spp. The range of spore sizes for *R. mengei* is unusual, about two-fold between smallest and largest (Fig. 1).

The only previously described *Rhizopogon* sp. close to *R. mengei* is *R. ventricisporus* A. H. Smith in Smith and Zeller (1966). However, *R. mengei* has regular, ellipsoid to oblong spores only rarely exceeding 14 x 6.5 µm and with an indistinct basidial attachment. In contrast, *R. ventricisporus* has highly variable spore shapes (ovate to fusoid-ventricose, subcylindric or variously elongated) and sizes (some up to 13-22 x 4-7 or rarely 22 x 4 µm) and a distinct, narrowly truncate basidial attachment ± 1.5 µm broad. *Rhizopogon guzmanii* Trappe & Cázares in Cázares *et al.* 1992) has spores in about the same size range as *R. mengei*, but it is in Section *Amylopogon* and has strongly amyloid spores.

Phylogenetic analysis of the internal transcribed spacer (ITS) and 28s regions of the nuclear ribosomal DNA support the placement of *R. mengei* in subsection *Rhizopogon* (L. Grubisha, unpublished data).

Rhizopogon spp. typically form mycorrhizae with species of Pinaceae. A. H. Smith was sent specimens collected "in duff under chinquapin oak," so he named it *R. quercicola* A. H. Smith in Smith and Zeller (1966). However, "chinquapin" is *Castanopsis chrysophylla*, a species in the Fagaceae that almost always grows in the understory of species of the Pinaceae. Consequently, association of *R. quercicola* with chinquapin cannot be confirmed by casual field observation. Indeed, *R. quercicola* is in section *Villosuli,* all species of which appear to be associated with *Pseudotsuga menziesii* (Mirb.) Franco. In the case of *R. mengei,* however, a thorough search revealed only *Adenostoma fasciculatum* (Rosaceae) at the holotype locality and no Pinaceae closer than ca 1 km. No Pinaceae were nearby at the Marin County locality, either. Studies of the *Adenostoma* mycorrhizae will be reported elsewhere.

Some specimens of *Rhizopogon mengei* had been partly eaten by small animals, presumably rodents.

ACKNOWLEDGMENTS

This study was supported in part by the Department of Energy Program for Ecosytem Research and the USDA Forest Service, Pacific Northwest Research Station.

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MYCOTAXON

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TWO NEW SPECIES OF DIDYMIUM (PHYSARALES, MYXOMYCETES) FROM JAPAN[*]

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Abstract: Two new species of *Didymium, D. laccatipes* and *D. angularisporum*, are described on the basis of collections from Japan. *Didymium laccatipes* resembles *D. squamulosum* in the general appearance of sporocarps but differs in having a glossy white stalk filled with rhombic lime crystals and strongly warted spores. *Didymium angularisporum* can be distinguished from all previously reported species of *Didymium* on the basis of having polyhedral, angular, smooth spores and a relatively rigid and thick brown capillitium.

Key words: Myxomycetes, Physarales, *Didymium*, Japan, taxonomy.

Introduction

The genus *Didymium* was established by Schrader (1797). Since then, about seventy species have been recognized in the genus. These species have traditionally been classified mainly on the basis of characters associated with exterior of the fruiting body and the contents of the capitulum (i.e., spore, capillitium, peridium, and columella). In our studies, we have observed longitudinal and transverse sections of fruiting bodies and, based upon these observations, have proposed a number of taxonomical revisions within the genus *Didymium* (Matsumoto 1996).

In the course of our taxonomic studies on the Japanese species of *Didymium*, two of the species we have collected are considered to be new to science. The first of these is a species most closely related to *Didymium squamulosum* (Alb. et Schw.) Fr., as described in Martin and Alexopoulos (1969), but it is clearly distinguished from *D. squamulosum* by the structure of the stalk. The second is a species that can be distinguished from all previously described species of *Didymium* on the basis of characters of the spore and capillitium. The stalk structure of this species suggests a close affinity to *D. melanospermum* (Pers.) T. Macbr. and *D. minus* (Lister) Morgan.

In the present paper, these two new species are described, with explanatory photomicrographs provided to supplement the descriptions. Possible relationships between the new species and what appear to be their closest relatives also are discussed.

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[*] Contribution from the Laboratory of Plant Taxonomy and Ecology, Hiroshima University. N. Ser. No.477.

Materials and methods

Observations of material were carried out with the use of light (LM) and scanning electron (SEM) microscopes. In addition to general LM observations, sections of sporocarps were examined. These sections were produced in the following manner: (1) each sporocarp was immersed in a drop of colorless nail polish on a glass slide, (2) after being air-dried, the sporocarp was hand-sectioned with a razor blade under a stereomicroscope, (3) the sections were transferred to separate slides and soaked in absolute alcohol, and (4) just before complete evaporation of the alcohol, the sections were mounted in Hoyer's medium. When it was necessary to remove lime from the fruiting body, the sections were treated with a dilute aqueous solution (1 N) of HCl before soaking in alcohol. For observation of the interior of the capitulum, nail polish and spores were removed by placing the sections in acetone before soaking in alcohol. For SEM observation, the air-dried samples were mounted on brass stubs with double-sided adhesive carbon tape, sputter-coated under vacuum with a film of gold in a JFC-1100 (JEOL) ion sputter, and examined with a JSM-25SIII (JEOL) scanning electron microscope at 12.5-20 kV accelerating voltage.

In addition to Japanese specimens of *Didymium* deposited in TNS and HIRO, type specimens of *Didymium floccosum* Martin *et al.* (*K. S. Thind 250,* isotype loaned from BPI) and *D. intermedium* Shroet. (*E. Ule 804,* holotype, obtained from BM) also were examined for comparison.

Morphological terminology follows Martin and Alexopoulos (1969) except for two terms proposed by Dörfelt and Marx (1991). As such, a sporangiate fruiting body is termed a "sporocarp" and a sporangial head of a stalked sporocarp is referred to as a "capitulum".

Taxonomy

Didymium laccatipes **J. Matsumoto, sp. nov.** (Figs. 1 - 9, 18 - 20)

Sporocarpia gregaria, stipitata, 0.7-1.0 mm alta. Capitulum depressiglobosum, infra cum umbilico, albicinereum, 0.4-0.6 mm diam., 0.2-0.4 mm crassum. Peridium membranaceum, tectum cum crystallis calcareis albis stelliformibus, purpureobrunneum vel pallide ochraceum, cum reticulis incoloratis. Hypothallus membranaceus. Stipes sursum attenuatus vel cylindricus, cum pagina laevi, cum costis paucis, intra completus cum calcibus crystallinis, 0.3-0.6 mm altus, laccatus, virgineus vel pallide ochraceus. Columella conica vel tholiformis, intra completa cum calcidus, alba vel pallide ochracea, 0.1-0.2 mm diam., 0.1-0.2 mm crassa. Capillitium abundum, gracile, rare ramosum, rare anastomosans, incoloratum vel purpureobrunneum. Sporae pallide brunneae, globosae, verrucosae, 9-11 μm diam. Plasmodium ignotum.

Figures 1-9, *Didymium laccatipes* J. Matsumoto (*J. Matsumoto150* - holotype).
1, sporocarp. 2, sporocarp showing columella. 3, longitudinal section of sporocarp. 4, longitudinal section of basal portion of stalk showing absence of dark refuse matter. 5 and 7, cross sections of stalks (5, intact section; 7, section treated with HCl). 6, fragment of peridium treated with dilute aqueous solution of HCl, showing dark (purplish brown) part and pale (pale ochraceous) part on same peridium. 8, longitudinal section of capitulum showing delicate capillitial elements. 9, spores. Bars: figs. 1 and 2 = 1 mm; figs. 3, 4, 5, 6, 7 and 8 = 100 μm; fig. 9 = 10 μm.

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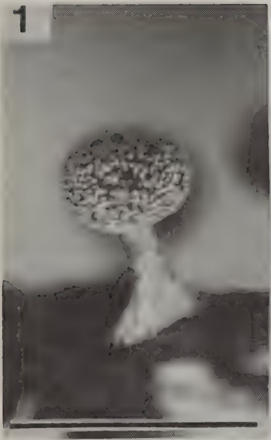

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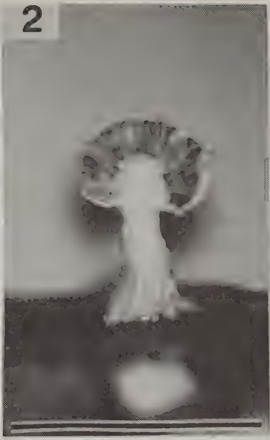

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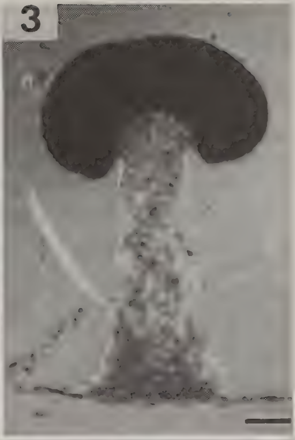

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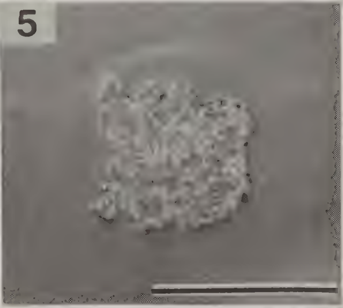

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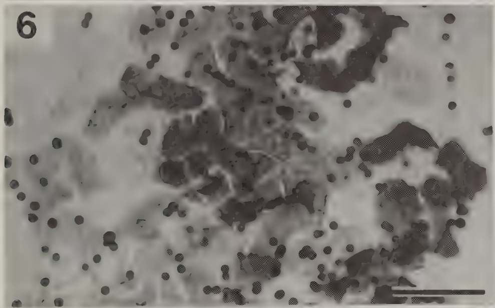

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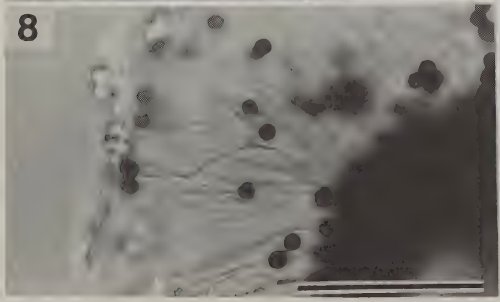

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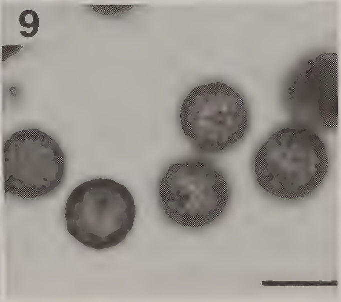

Fructification sporocarpous. Sporocarps gregarious, stalked, 0.7-1.0 mm high. Capitulum depressed-globose to discoid, umbilicate below, grayish white, 0.4-0.6 mm in diam., 0.2-0.4 mm thick. Peridium membranous, covered with white stellate lime crystals, purplish brown to pale ochraceous, with colorless reticulation, dehiscing from above, persistent in the lower part. Hypothallus membranous, colorless to very pale ochraceous, containing lime crystals only at the stalk base, often connecting sporocarps as calcareous, vein-like ridges. Stalk slightly tapering upward, nearly cylindrical, almost smooth, with few delicate, slightly wavy costae on surface, filled with rhombic lime crystals throughout, not containing refuse matter, glossy, pure white to very pale ochraceous, 0.3-0.6 mm high; the apex inserted through the umbilicus into the capitulum. Columella conical to dome-shaped, containing lime crystals, white to very pale ochraceous, with reticulate ridges on surface, 0.1-0.2 mm in diam., 0.1-0.2 mm thick. Capillitium relatively abundant, delicate, sparsely branched, hardly anastomosing, radiating from reticulate ridges of the columella to the peridium, colorless to very pale purplish brown. Spores dark brown in mass, pale brown in transmitted light, globose, strongly warted, often with dark clusters of warts, 9-11 μm in diam.

Etymology: From the Latin, laccatus (vanished) + -pes (-footed), in reference to the glossy stalk appearing to be varnished.

Specimens examined: Japan, Wakayama-ken, Nishimuro-gun, Shirahama-cho, on fallen leaves of *Daphniphyllum* sp., 24 July, 1990, leg. J. Matsumoto, *Matsumoto 150* (holotype in HIRO, isotype in TNS). Japan, Fukui-ken, Tsuruga-shi, Tedutu-yama, about 50 m alt., on fallen leaf of *Illicium religiosum*, 6 Aug., 1983, leg. J. Matsumoto, *Matsumoto 151* (HIRO). Japan, Nara-ken, Kashihara-shi, Kashihara-jingu, on fallen leaf (unknown broad-leaved tree species in forest of *Castanopsis*), 9 July, 1995, leg. H. Kobayashi, det. J. Matsumoto, *Matsumoto 1652* (HIRO).

Habitat: On fallen leaves.

Distribution: Japan (Honshu: Fukui-ken, Nara-ken, Wakayama-ken).

Season of fructification: July and August.

The general appearance of sporocarps of *Didymium laccatipes* is similar to that of *D. squamulosum*. However, the new species can be distinguished from the latter by the stalk structure. The stalk of *D. laccatipes* is filled with rhombic lime crystals and is not frosted with lime on its external surface (Figs. 3, 5, 7 and 20). The stalk of *D. squamulosum* is not filled with lime inside but is covered with lime on its external surface.

Didymium laccatipes is also distinguishable from *D. squamulosum* by the conical to dome-shaped columella, the pale ochraceous to pale purplish brown peridium, and the more prominently warted spore (Figs. 2, 6, 9, 18 and 19).

Didymium laccatipes shows a close affinity to *D. floccosum* and *D. intermedium* in the stalk structure. These three species all have a stalk with the features described above. However, *D. laccatipes* is easily distinguishable from the latter two species by a combination of other features (see Table 1).

Didymium floccosum differs from *D. laccatipes* in the ochraceous stalk containing refuse matter in its basal part, the dark clavate columella, and the somewhat more slender sporocarps. *Didymium intermedium* differs from *D. laccatipes* in the colorless peridium and spores covered with longer spines that are arranged to form a partially banded reticulation.

Table 1. Comparison of sporocarpous characteristics of *Didymium intermedium, D. laccatipes* and *D. floccosum*

	D. intermedium (*E. Ule 804* in BM)	*D. laccatipes* (*Matsumoto 150* in HIRO)	*D. floccosum* (*K. S. Thind 250* in BPI)
Capitulum shape	hemispheric to depressed-globose	depressed-globose	depressed-globose to globose
Peridial color	colorless	pale ochraceous to purplish brown	purplish brown
Columella shape; color	discoid to dome-shaped or inconspicuous; white	conical to dome-shaped; white	clavate; dark brown
Stalk color	pure white to pale ochraceous	pure white to pale ochraceous	ochraceous, darker in the basal portion which contains refuse matter
Capillitium	profuse, delicate	profuse, delicate	profuse, delicate
Spore ornamentation; size	spines arranged to form a distinct reticulation; 8-11 μm	strongly warted often with dark clusters of warts; 9-11 μm	densely verrucose, the warts often clustered and arranged in an obscurely reticulate pattern; 8-10 μm

The morphological data for *Didymium intermedium* and *D. floccosum* are from our own observation of their type specimens and their original descriptions (Hennings 1896; Martin *et al*. 1959).

Didymium angularisporum **J. Matsumoto, sp. nov.** (Figs. 10 - 17, 21 - 23)

Sporocarpia gregaria, stipitata, 0.7-1.0 mm alta. Capitulum depressiglobosum, infra cum umbilico, albicinereum, 0.4-0.5 mm diam., 0.3-0.4 mm crassum. Peridium membranaceum, tectum cum crystallis calcareis albis stelliformibus, purpureobrunneum, cum reticulis incoloratis. Hypothallus membranaceus. Stipes sursum attenuatus vel cylindricus, cum costis numerosis sinuosis pellucidis, intra completus cum residuo furvo, 0.4-0.6 mm altus, atro-brunneus vel brunneorus. Columella disciformis, intra completa cum calcidus, atro-brunnea vel nigra, ca. 0.3 mm diam., ca. 0.2 mm crassa. Capillitium rigidulum, rare ramosum praeter extremites, cum extremitibus ramosis, rare anastomosans, purpureobrunneum vel brunneum. Sporae purpureobrunneae, polyedricae, angulares, laeves, 9.5-11 μm diam. Plasmodium ignotum.

Fructification sporocarpous. Sporocarps gregarious, stalked, 0.7-1.0 mm high. Capitulum depressed-globose, umbilicate below, whitish gray, 0.4-0.5 mm in diam., 0.3-0.4 mm thick. Peridium membranous, covered with relatively small, white stellate lime crystals, purplish brown, with colorless reticulation only on the upper part, dehiscing from above. Hypothallus membranous, containing refuse matter, brown at the stalk base, connecting sporocarps as a membranous sheet, not calcareous. Stalk tapering upward, nearly cylindrical, filled with refuse matter from the base nearly to the apex, brown to dark brown in transmitted light, with many wavy translucent costae, 0.4-0.6 mm high, expanded like a calyculus at its apex; the apex inserted through the umbilicus into the capitulum. Columella discoid, containing lime crystals, dark brown to black, covered with reticulate ridges on surface, 0.3 mm in diam., 0.2 mm thick. Capillitium relatively rigid and thick, almost unbranched except for at the extremities which branch dichotomously, hardly anastomosing, radiating from the surface reticulate ridges of columella to the peridium, brown, nearly colorless at the extremities. Spores dark brown in mass, purplish brown in transmitted light, polyhedral, angular, almost smooth, 9.5-11 μm in diam.

Etymology: From the Latin, angularis (angular) + -sporus (-spored), in reference to the polyhedral angular spore.

Specimen examined: Japan, Yamagata-ken, Higashi-okitama-gun, Kawanishi-cho, Tamaniwa, on dead herbaceous leaves and stems, 14 Oct., 1983, leg. H. Hagiwara, *M-H-3526* (holotype in TNS, isotype in HIRO).

Habitat: On dead herbaceous leaves and stems.

Distribution: Japan (Honshu: Yamagata-ken).

Season of fructification: October.

Didymium angularisporum is characterized by the polyhedral, angular spores and the relatively rigid and thick brown capillitium (Figs. 14, 17 and 21). The spores appear smooth under LM but are very minutely warted under SEM (Figs. 22 and 23).

Didymium angularisporum is similar to *D. minus* and *D. melanospermum* var. *melanospermum* in the dark brown stalk filled with refuse matter, the dark brown columella, and the purplish brown peridium (Figs. 11, 13, 15 and 16). *Didymium minus* can be distinguished from *D. angularisporum* by the warted globose spores with dark clusters of warts, the delicate pale capillitium, and the subglobose columella. *Didymium melanospermum* var. *melanospermum* possesses a thick brown capillitium, but the latter is more abundant and sinuous than that of *D. angularisporum*. *Didymium melanospermum* var. *melanospermum* also differs from *D. angularisporum* in the strongly warted, globose spores and the shorter stalk that is often immersed in the umbilicus of the capitulum.

Acknowledgements

We thank Dr. S. L. Stephenson, Fairmont State College, Fairmont, West Virginia, USA, and Dr. H. Hagiwara, National Science Museum, Tsukuba, Ibaraki-ken, Japan, for critical reviews on the manuscript, and Mr. H. Kobayashi, Kyoto University, Kyoto, Japan, for offering a useful specimen. Thanks are also due to the directors and curators

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Figures 10-17, *Didymium angularisporum* J. Matsumoto (M-H-3526 - holotype). 10, sporocarp. 11, sporocarp showing columella. 12, longitudinal section of sporocarp. 13, cross section of stalk. 14, longitudinal section of capitulum showing capillitial elements. 15, longitudinal section of stalk (note that the stalk contains dark colored refuse matter throughout). 16, fragment of peridium. 17, spores. Bars: figs. 10 and 11 = 1 mm; figs. 12, 13, 14, 15 and 16 = 100 μm; fig. 17 = 10 μm.

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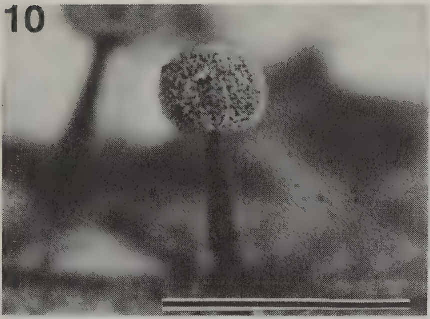

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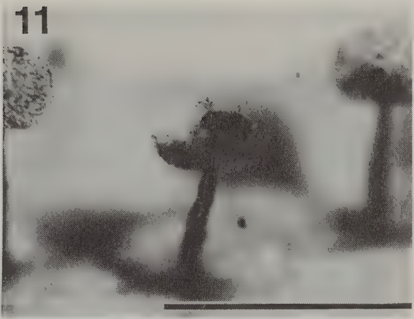

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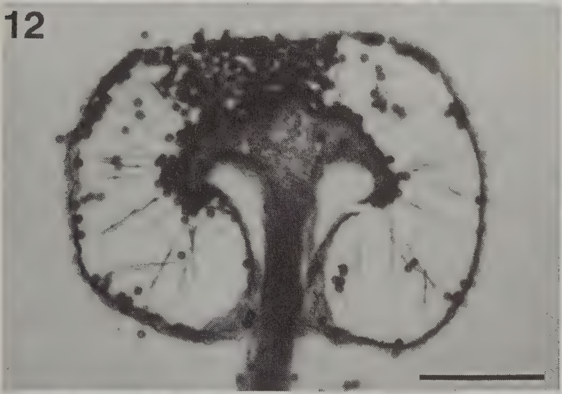

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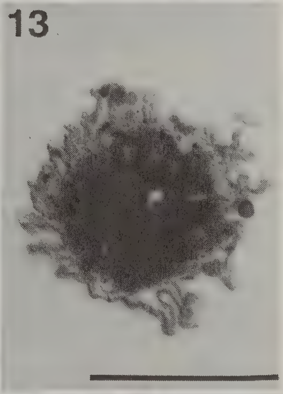

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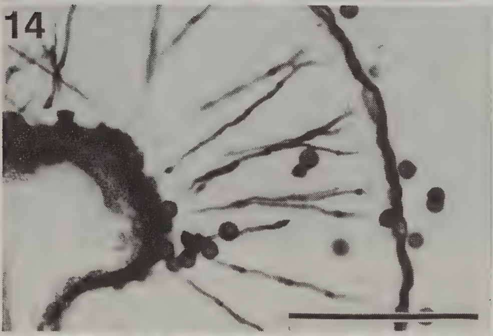

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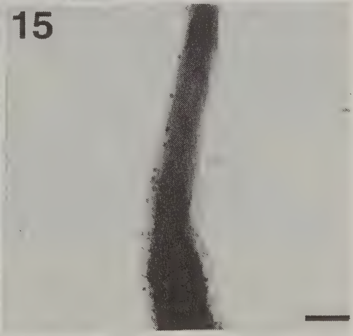

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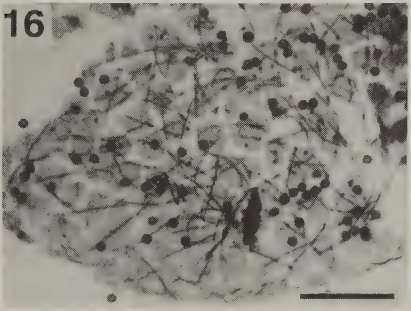

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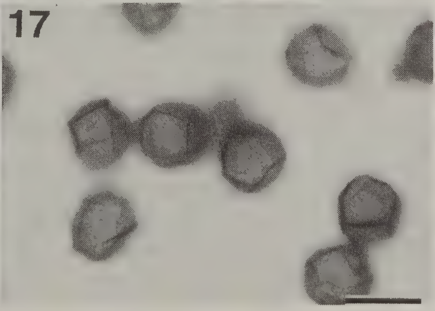
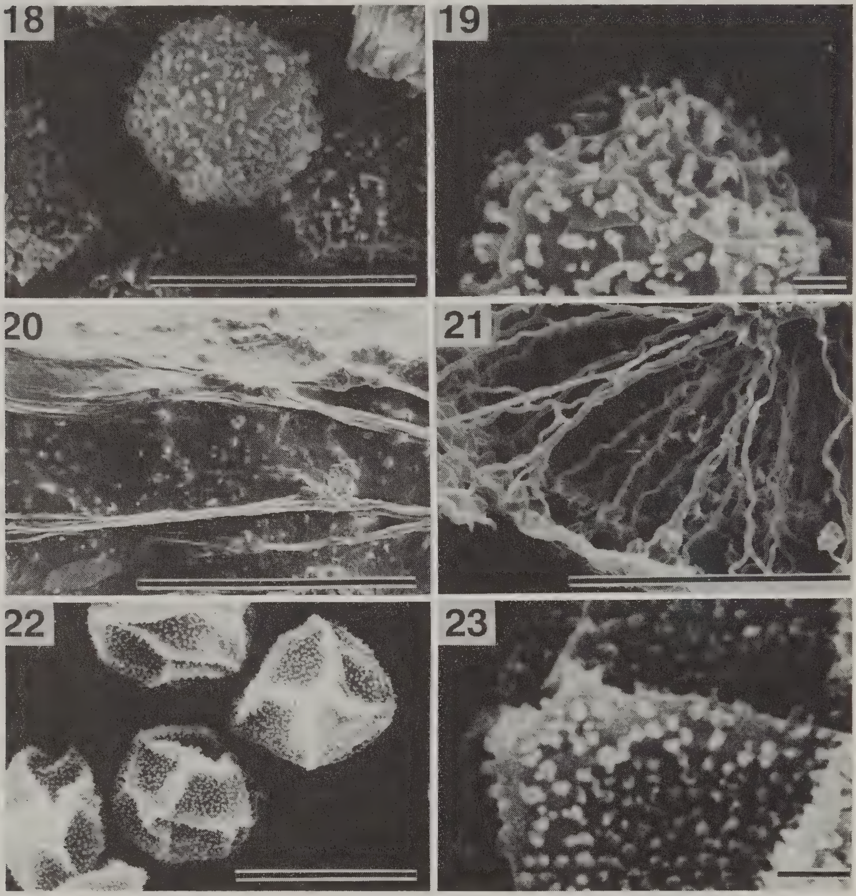


of the Natural History Museum, London (BM), U. S. National Fungus Collections, Beltsville (BPI) and National Science Museum, Tokyo (TNS) for loaning specimens.



Figures 18-23, Photomicrographs by SEM. 18-20, *Didymium laccatipes* J. Matsumoto (18, spores; 19, part of spore; 20, surface of stalk). 21-23, *Didymium angularisporum* J. Matsumoto (21, part of capitulum showing capillitial elements; 22, spores; 23, part of spore). All photomicrographs from holotype. Bars: figs. 18, 20, 21 and 22 = 10 μm; figs. 19 and 23 = 1 μm.

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MYCOTAXON

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A NEW SPECIES OF CANOPARMELIA FROM NORTH AMERICA (LICHENIZED ASCOMYCOTINA, PARMELIACEAE)

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ABSTRACT : *Canoparmelia amabilis* Heiman & Elix from eastern North America is described as new to science.

Introduction

During the course of a lichen inventory project in the Obed Wild and Scenic River Corridor in Tennessee, USA, we encountered a species of *Canoparmelia* which had features which were inconsistent with the known species of this genus (Elix *et al.* 1986; Elix 1994), so we are describing it as new. Throughout the present work chemical constituents were identified by thin layer chromatography (Culberson 1972; Culberson, Culberson & Johnson 1981; Culberson & Johnson 1982; Elix & Ernst-Russell 1993; Elix *et al.* 1988), high performance liquid chromatography (Feige *et al.* 1993) and comparison with authentic samples.

Canoparmelia amabilis Heiman & Elix, sp. nov. Fig. 1

Species cum thallo ut in *Canoparmelia caroliniana* sed ab hac specie isidiis latioribus, coralloidibus et colore pallido subto differt.

Type: United States of America. Tennessee, Morgan Co., Obed Wild and Scenic River Park, knife edge ridge above Clear Creek Junction, on exposed sandstone rock, *Karin Heiman 90015*, 7 October 1996; holo: US.

Thallus saxicolous or corticolous, foliose, adnate to tightly adnate, to 3-8 cm wide. *Lobes* imbricate, sublinear to subirregular, irregularly branched, crenate or deeply incised, 2.0-5.0 mm wide, apices rotund. *Upper surface*

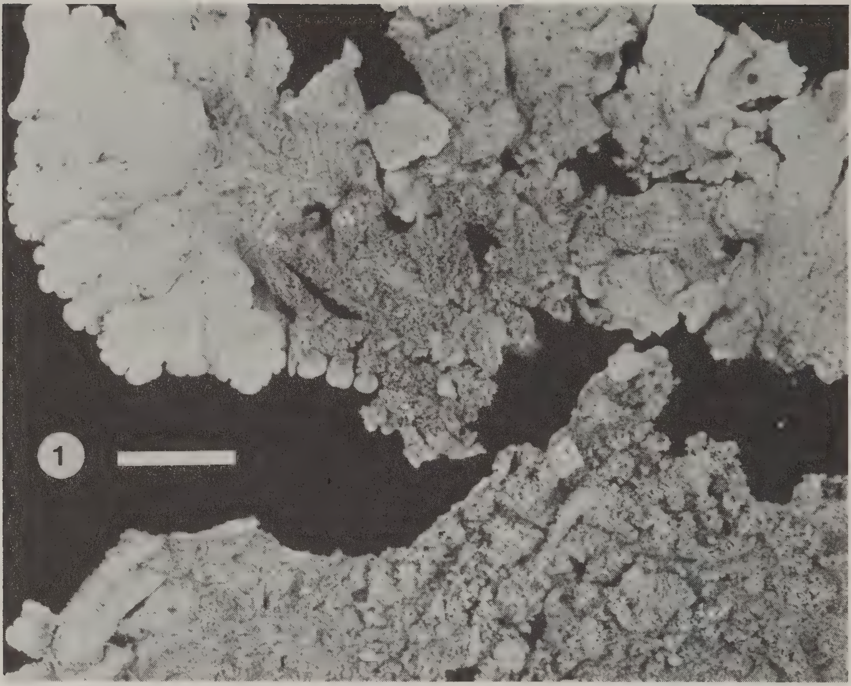

Figure 1. *Canoparmelia amabilis* (part *T. H. Nash 19814* in CANB). Scale bar = 5 mm.

pale gray to ashy white or gray-green, flat, smooth but becoming rugulose, white maculate, usually finely cracked, shiny at the apices, without soredia; densely isidiate, the isidia cylindrical, 1-3 mm high, becoming densely branched and coralloid; apices syncorticate, ±blackening. *Medulla* white. *Lower surface* pale brown to brown, with an ivory to pale tan marginal zone; rhizines moderately dense, simple or tufted, concolorous. *Apothecia* and *pycnidia* not seen.

CHEMISTRY. Cortex K+ yellow; medulla K-, C-, KC-, P-; containing atranorin (minor), chloroatranorin (minor), perlatolic acid (major), stenosporic acid (major), glomelliferic acid (minor), anziaic acid (trace), 4-*O*-demethylstenosporic acid (trace), divaricatic acid (trace).

In overall morphology this new species closely resembles *Canoparmelia caroliniana* (Nyl.) Elix & Hale, with both species having adnate thalli, relatively broad lobes with rotund apices, and a white maculate, isidiate upper surface. Although the two species are chemically identical, they differ in the color of the lower surface and in the nature of the isidia.

Canoparmelia caroliniana has a black to black-brown lower surface with a paler, brown marginal zone whereas *C. amabilis* has a pale brown to brown lower surface with an ivory to pale tan marginal zone. Further, the isidia are fragile, narrow (to 0.07 mm in diameter) and mainly simple in *C. caroliniana*, but are denser, more robust (to 0.12 mm in diameter) and densely branched to coralloid in *C. amabilis*.

This new species was first recognized in several collections from the Obed Wild and Scenic River National Recreation Area, which consists of 72 km of steep river gorges along the Obed River and its tributaries in the Cumberland Plateau, which is part of the larger Allegheny Plateau Province. The land, which is managed by the National Park Service, occurs in Morgan and Cumberland Counties, Tennessee, USA. The area is characterized by flat rolling plateaus dissected by sheer gorge walls where the rivers have cut through. The geology of the site is rather simple, generally composed of Pennsylvanian sandstones, with occasional exposures of shale or coal. (Wilson et al. 1956). Soils are generally acid, highly leached and nutrient poor. Natural communities on the ridges are quite xeric, dominated by oaks and *Pinus virginiana*, while the slopes and narrow creek beds are more mesic, with *Acer barbatum*, *A. rubrum*, *Betula nigra*, *Carpinus caroliniana*, *Carya* spp., *Fagus grandifolia*, *Ilex opaca*, *Liriodendron tulipifera*, *Pinus strobus* and *Tsuga canadensis* all common.
The North Carolina location mentioned below is located near the escarpment on the Blue Ridge Mountains. It seems likely that this new species will also be found between these locations.

Specimens Examined

UNITED STATES OF AMERICA. North Carolina, Polk Co., Glenwalden Circle in Tryon, 35°12'N, 82°14'30"W, 360 m, on *Acer rubrum* in deciduous forest, *T. H. Nash 19,814*, 27 December 1982 (CANB).
Tennessee, Morgan Co., Obed Wild and Scenic River Corridor, Lily Boulderfield Trail south of Lily Bridge, between road and river, 335 m, on shaded sandstone boulder in forest, *Karin Heiman 90106*, 6 October 1997 (US); Morgan County, Obed Wild and Scenic River Corridor, cascade area of Mill Creek, on bark of fallen tree, *Karin Heiman 90212*, 7 August 1997 (US).

ACKNOWLEDGEMENTS

We wish to thank Stuart Hay and Neal McCracken of the Photographic Unit, ANU, for preparing the photograph and Doug Verdon for checking the Latin description. This lichenological study would not have been initiated were it not for concern and funding by the US National Park Service and The Nature Conservancy, which is gratefully acknowledged. KH also wishes to acknowledge the assistance of Stephanie Wilds, Robert Emmott, Gregg Mattson, as well as Drs Paula DePriest, Bruce McCune, Roger Rosentreter and Bruce Ryan.

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MYCOTAXON

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THREE NEW SPECIES OF RUSSULA

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ABSTRACT

Descriptions and illustrations are presented of three new species of *Russula*: *R. avellaneiceps*, *R. glutinosa*, and *R. primaverna.*

INTRODUCTION

The number of *Russula* collections defying positive identification is disconcertingly high. Some may be only marginally different from a recognized species, or a tentative identification to one of the published names which were poorly or incompletely described in the early part of this century may often be made. Yet some collections will exhibit characteristics which are so strongly diagnostic that the taxon will be recognized as being indisputably different from known species. Several of those within this category are detailed here.

MATERIALS AND METHODS

Color terms and designations used in the following descriptions are cited from Maerz & Paul (1950), e.g., M&P 7L12, or from the ISCC-NBS system (Kelly & Judd, 1955), e.g., 54 brownish Orange. Otherwise, color names are the author's concept of general color hues. The massed spore color designated by a capital letter from A to H refers to the spore color code of Pearson (1950), or designated by a Roman numeral alphabetical combination, e.g., IVe, refers to Romagnesi's (1967) color code. Spores were examined mounted in Melzer's reagent, size determined in lateral view, with ornamentation excluded. Other microscopic descriptions were made from sections mounted in 3% KOH and 10% NH_4OH. Illustrations were made with the aid of a drawing tube. The pileus was studied from tangential sections. Gelatinous matrices on reconstituted material were best observed in NH_4OH.

Russula avellaneiceps Fatto, sp. nov. Figs. 1-5

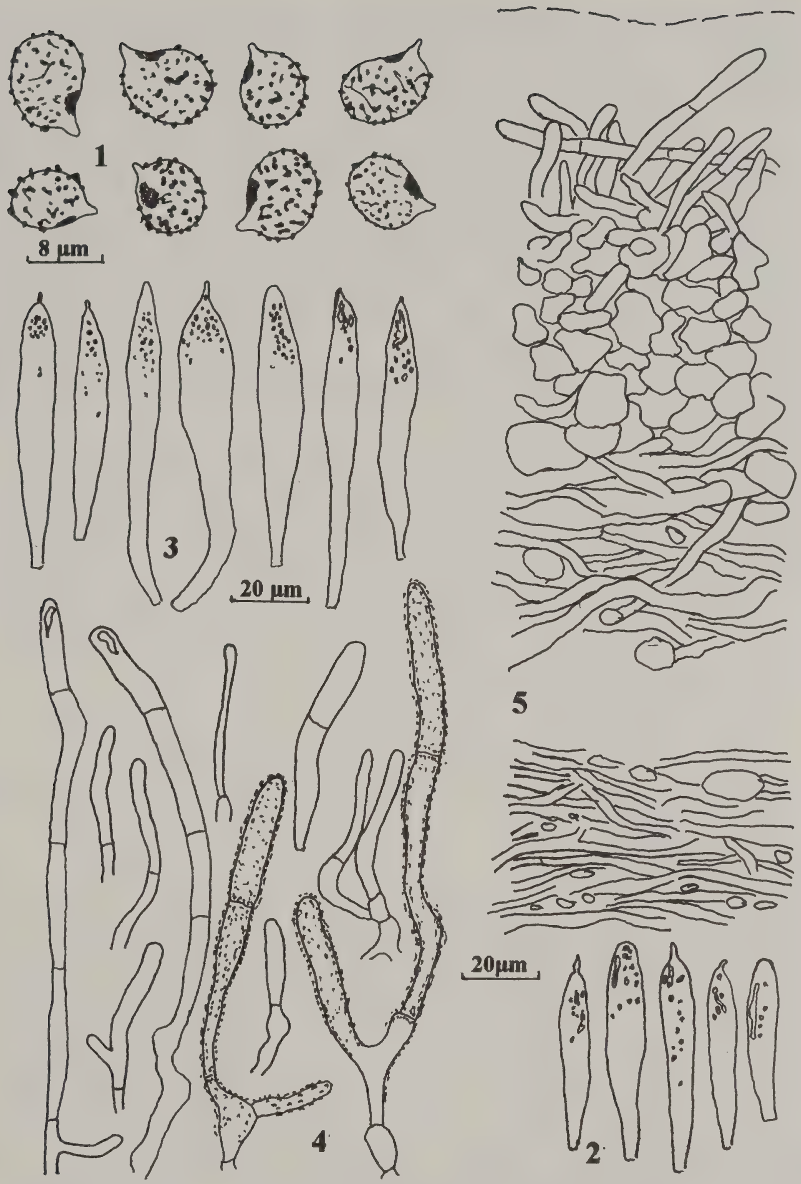
Pileus 4-8 cm latus, convexus, maturans ad plano-depressum; margo aequus, aetate leniter striatulus; cutis separabilis usque ad 2/3 partes radii, glabra, avellanea; trama alba, immutans; sapor mitis; odor nullus. Lamellae adnatae, confertae, ochrae. Stipes ad 8 x 3 cm, augens deorsum, demum cavus, glaber, roseus. Sporae ochraceae (Romagnesi IVe), 8-10.4 x 6.4-8 µm, flocculae 0.4-0.8 µm altae, segregatae vel cum paucis et brevibus connectivis. Cystidia hymenialia 42-88 x 8-14 µm, fusiformia, fusoidea-clavata, cum accumulatis parvis et reflexis in sectione apicali, aliter vacua. Pileus subcutis habens hyphus libratas-contextas filiformes ad hyphas inflatas, formantes pseudoparenchymatosam zonam in parte superiori. Pileus epicutis desidens in matrice gelatinosa, fines hypharum procumbentes, ascendentes ad erectas, apices rotundi; etiam cum multiseptatis incrustatis originalibus hyphis; sine pileocystidiis. Stipes cutis cum hyphis similibus epicutici pilei, sine SV+ hyphis.

Holotypus: Lectus a Debbie Johnson, NAMA 97-210 (Fatto sp101), sub pinus contorta prope Dillon Reservoir, Summit County, Colorado, USA. 15 August 1997. Conservatus in herbario Chicago Field Museum of Natural History (F).

Pileus 4-8 cm broad, convex, maturing plano-depressed; margin even, in age becoming slightly striate 2-4 mm inward from the edge; cuticle viscid when wet, dull dry, disc sometimes shiny, glabrous, separable to 1/2-2/3 the radius, brownish hues reminiscent of a filbert, usually 54 brownish Orange, M&P 14A12, 13C12, to 13E9, the disc sometimes darker, to 55 strong Brown, M&P 7L12, sometimes sun bleached in areas as light as M&P 11I4 or roughly 73 pale Orange Yellow; trama thin, to 3 mm at mid-radius, brittle, white, unchanging when injured, with a mild taste and no significant odor. **Lamellae** to 8 mm broad at mid-radius, rounded in front, adnate to adnexed, close, some basal forking, a few long lamellulae, edges even, brittle, taste mild, pale ochre becoming deep ochre. **Stipe** 4-8 x 1-3 cm, subequal or enlarging toward the base, dry, glabrous, longitudinally rugulose, whitish with at least half of the surface washed with 5 moderate Pink, M&P 3F1, stuffed, becoming hollow, unchanged when bruised.

Chemical reactions (stipe surface and trama): sulphovanillin (SV) nil; 10% $FeSO_4$ nil to faint salmon-tan; 2% phenol slowly to a light and finally dark brown; formalin colorless; guaiac light grayish blue.

Spores ochraceous in mass, H or IVe, 8-10.4 x 6.4-8.0 µm, subglobose, obovate, elliptic; ornamentation of amyloid warts 0.4-0.8 µm high, mostly isolated, connate or with a few short connectives; suprahilar area a small lightly amyloid patch; hilar appendix inamyloid, conspicuous. **Basidia** 40-66 x 11-14 µm, narrowly clavate, 4-sterigmate. **Pleurocystidia** 60-88 x 8-14 µm, subcylindric, fusiform, fusoid-clavate, apices rounded, acute, mucronate or short-appendiculate, empty or with refractive granules loosely



FIGS. 1-5. *Russula avellaneiceps* (NAMA 97-210, holotype). 1. Spores. 2. Cheilocystidia. 3. Pleurocystidia. 4. Hyphal ends from pileus cuticle. Incrusted hyphae as observed in SV. 5. Pileus cuticle (dotted line represents surface of gelatinous matrix). Approximately 1/3 of subcutis not illustrated. Outlined areas in hyphae and cystidia indicate refractive masses.

congregated in the apical section, these granules refractive-grayish in SV, arising from the subhymenium or outer edge of the trama, projecting to 28 µm beyond the basidioles, scattered. **Cheilocystidia** 42-62 x 8-10 µm, fusoid-clavate, clavate, apices rounded, short-appendiculate, empty or with refractive granules as in the pleurocystidia. **Subhymenium** 32-60 µm thick, pseudoparenchymatous to occasionally prosenchymatous in the lower region. **Trama** without or with rare SV-, smooth vascular hyphae. **Pileus subcutis** 160-280 µm thick, horizontal-interwoven to ascending, hyaline, filamentous to inflated hyphae 2-16 µm wide, at mid-radius the upper portion, approximately one third, consists of inflated cells forming a pseudoparenchymatous layer, cells 5-30 µm wide, whereas, closer to the margin, at about one fifth of the radius, only an occasional inflated cell exists among the filamentous hyphae; also with common smooth vascular hyphae 4-10 µm wide. **Pileus epicutis** 40-70 µm thick, embedded in a gelatinous matrix, hyaline, repent, ascending to erect hyphae 2-6 (-8) µm wide, apices rounded, and common primordial hyphae 3-6 (-8) µm wide, to 160 µm long, multiseptate, incrusted with refractive-grayish granules when observed in SV, an occasional terminal segment containing a small refractive mass; without pileocystidia. **Stipe cuticle** of longitudinal-interwoven, nongelatinous hyphae 2-6 µm wide, with tips projecting to 60 µm, and incrusted primordial hyphae 4-8 µm wide, extending to 130 µm, septate, apices rounded, sometimes attenuated, occasionally with a small refractive mass in the terminal segment, incrusting granules and refractive mass are refractive-grayish in SV; also with smooth vascular hyphae 2-10 µm wide, SV-, common.

Habitat solitary on soil under lodgepole pine (*Pinus contorta* Dougl.); August.

MATERIAL EXAMINED: COLORADO: Summit Co.: Prospector Campground, Dillon Reservoir, Debbie Johnson, 15 Aug 1997, NAMA 97-210 (Fatto sp101) (Holotype, F; Isotype, NY); Peninsula Recreation Area east of Dillon Reservoir, Nathan Wilson, 14 Aug 1997, Fatto 1039 (Paratype, NY).

OBSERVATIONS: *Russula avellaneiceps* is placed in Singer's (1986) subsection *Amethystinae* of section *Russula* because of the rich yellow spore print, mild taste, medium stature, unchanging trama, coniferous habitat, hymenial cystidia with only sparse apical SV+ content, primordial hyphae on the pileus and stipe cuticles, and the absence of pileocystidia, caulocystidia and any SV+ hyphae. While no significant odor was noted on the lamellae or pileus, the stipe base was not investigated for the presence of any iodoform odor. This taxon was collected at the 1997 North American Mycological Association Foray at Copper Mountain, CO. Once the original specimen was studied, additional specimens were readily recognized by the distinctive hazelnut pileus color, pink stipe, ochre lamellae, mild taste, and a quick microscopic inspection of a crush mount in SV of the pileus cuticle and lamellae to observe the primordial hyphae and hymenial cystidia.

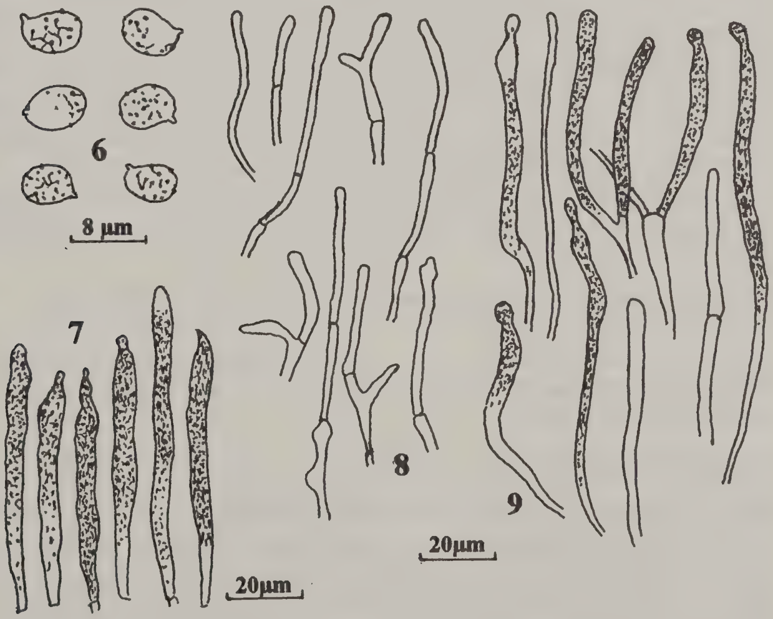
Russula glutinosa Fatto, sp. nov. Figs. 6-9

Pileus 6-9 cm latus, convexus, maturans ad plano-depressum; margo aequus; cutis

glutinosa, resiliens, separabilis usque ad 1/3 partem radii, albida ad pallide ochraceam, media cutis leviter luteo-fusca; trama dura, immutans. Lamellae adnatae, subdistantes, cum abundantibus lamellulis, cremeis, immutantibus, sapor acer. Stipes ad 6 x 2 cm, cremeus, glaber, firmus, immutans. Sporae cremeae (Romagnesi IIc), 5-7 x 4-5 µm, flocculae 0.1 µm altae, segregatae aut cum paucis gracilibus connectivis. Cystidia hymenialia 60-85 x 4-6 µm, abundantia, completa granulis reflexis-griseolis. Pileus cutis ad 300 µm crassam, desidens in matrice gelatinosa, extendens usque ad 280 µm supra extremas hyphas; pileus subcutis habens hyphas libratas-contextas 2-4 µm latas; pileus epicutis habens trichodermis hypharum hyalinarum 2-4 µm lata, sine pileocystidiis. Hyphae stipitis cuticis similes cutici pilei, sed cum multis oleiferis extremis hyphis 3-5 µm latis cum reflexo-griseolo contento.

Holotypus: Lectus a R. M. Fatto 1034, in Mendham town park, Morris County, New Jersey, USA. 6 August 1997. Conservatus in herbario New York Botanical Garden (NY).

Pileus 6-9 cm broad, convex, aging plano-depressed; margin even; cuticle glutinous, shiny, rubbery, separable 1/3 or slightly more of the radius, whitish or pale ochraceous, a light tan centrally, fairly uniformly 76 light yellowish Brown, M&P 12E7 when dried; trama 5 mm thick at mid-radius, firm to hard, whitish, unchanging when bruised, with a weak definite but nondescript odor. **Lamellae** 4 mm broad at mid-radius, narrowed in



FIGS. 6-9. *Russula glutinosa* (Fatto 1034, holotype). 6. Spores. 7. Hymenial cystidia. 8. Hyphal ends from pileus cuticle. 9. Hyphal ends from stipe cuticle.

front, adnate, subdistant, few basal forks, lamellulae arranged in three tiers, edges even, flexible, taste acrid, creamy with pale brownish-salmon stain spots, unchanging when bruised. **Stipe** 4-6 x 1.5-2 cm, equal to slightly enlarging basally, moist to viscid, glabrous, longitudinally rugulose, creamy, solid, firm, unchanging when bruised but slowly developing tan handling stains.

Chemical reactions (stipe surface and trama): SV nil; 10% $FeSO_4$ nil to faint salmon-pink; 2% phenol slowly to medium brown; formalin nil; guaiac dull greenish blue.

Spores pale cream in mass, C-D or IIc, 5-7 x 4-5 µm, obovate, elliptic; ornamentation of amyloid warts 0-0.1 (-0.2) µm high, isolated or with indistinct fine lines rarely forming any closures; suprahilar area a plage; hilar appendix inamyloid, inconspicuous. **Basidia** 28-45 x 5-7 µm, subclavate, usually 4-sterigmate, occasionally 2-sterigmate. **Hymenial cystidia** 60-85 x 4-6 µm, subcylindric, somewhat flexuous, apices rounded, acute, capitellate, filled with refractive-grayish granular content, moderately SV+, arising in the subhymenium or at the outer edge of the trama, embedded or projecting to 6 µm beyond the basidioles, abundant. **Subhymenium** 40-64 µm thick, prosenchymatous, hyphae 2-5.6 µm wide, closely septate. **Trama** without or with rare oleiferous hyphae and with rare smooth vascular hyphae. **Pileus subcutis** 120-180 µm thick, colorless in mass, horizontal-interwoven, hyaline hyphae 2-4 µm wide; also with smooth vascular hyphae 3-12 µm wide and rare oleiferous hyphae. **Pileus epicutis** 80-120 µm thick, pale buff in mass, embedded in a gelatinous matrix which extends to 280 µm beyond the hyphal tips, a trichoderm of widely septate, hyaline, occasionally branched, filamentous hyphae 2-4 µm wide, apices rounded and occasionally subcapitate, without pileocystidia. **Stipe cuticle** 120-220 µm thick, embedded in a thin gelatinous matrix, of interwoven, repent to ascending, hyaline hyphae 2-4 µm wide, and numerous irregularly constricted, subcylindric caulocystidia 3-5 µm wide, containing refractive-grayish granules, SV-, apices rounded, subcapitate to appendiculate, which may be ends of oleiferous hyphae or arise from the connective hyphae; also with numerous smooth vascular hyphae 4-12 µm wide.

Habitat scattered on soil in mixed beech, oak, deciduous woods; July, August.

MATERIAL EXAMINED: NEW JERSEY: Morris Co.: Mendham town park, 10 Jul 1994, Fatto 798; 6 Aug 1997, Fatto 1034 (Holotype, NY); 2 Aug 1998, Fatto 1142. Passaic Co.: Hewitt (Jennings Creek), Aaron Norarevian, 28 Jul 1996, Fatto 982.

OBSERVATIONS: *Russula glutinosa* is placed in subsection *Lactarioideae* Maire of section *Compactae* (Fr.) Maire as detailed by Shaffer (1964), which essentially corresponds to Singer's section *Plorantes* (Bat.) Singer. Macroscopic diagnostic features are the pallid coloration, firm trama, abundant tiered lamellulae, absence of graying on bruising or aging, acrid taste, nil formalin reaction, and cream spore coloration. The rubbery, glutinous pileus cuticle is species specific. Microscopically, the abundant SV+ hymenial cystidia and the small, almost smooth spores are prominent characteristics. The absence of a distinct suprahilar amyloid spot as indicated for Singer's section *Plorantes* is not considered significant.

Russula primaverna Fatto, sp. nov. Figs. 10-13

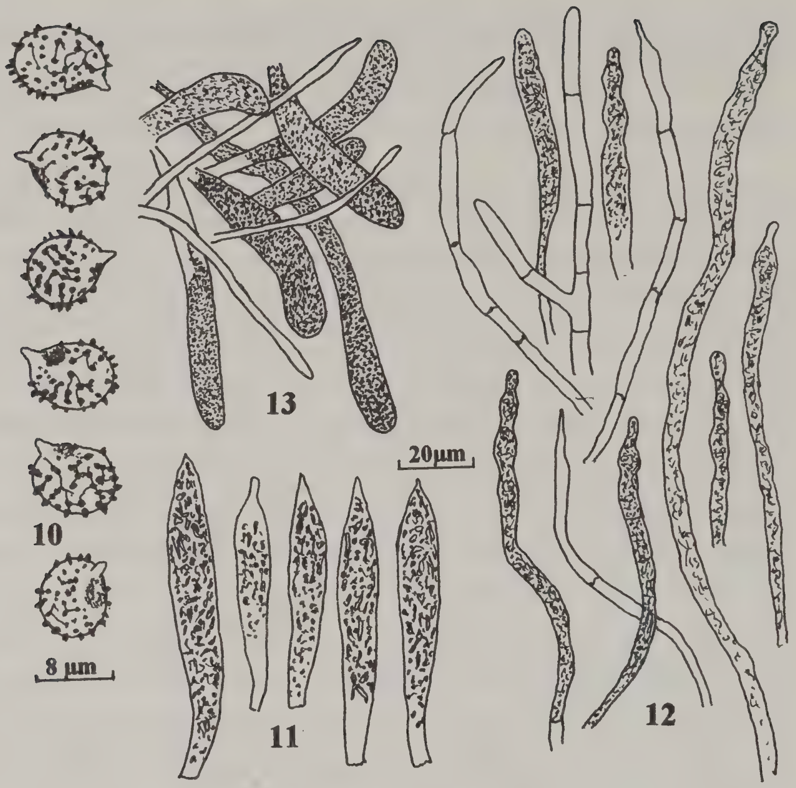
Pileus 3-9 cm latus; margo leniter striatulus; cutis separbilis usque ad dimidium radii, vernicosa, summa medio-brunnea; trama alba, immutans; sapor acer. Lamellae subacutae in fronte, adnatae, medio-propinquae, albae. Stipes ad 6.5 x 2.5 cm, aequus, albus, immutans. Sporae albae abundanter, 8.0-10.0 x 6.0-7.2 µm, flocculae 0.5-1.0 µm altae, cum gracilibus connectivis sed nullo reticulo. Cystidia hymenialia 56-84 x 8-12 µm, fusoidea, completa, abundantia. Pileus subcutis habens hyphas hyalinas, libratas-contextas. Pileus epicutis habens ixotrichodermis hypharum hyalinarum filimentosarum atque abundantia et torta et subcylindrica pileocystidia oleiferorum hyphalium finium. Stipes cutis cum contextis, hyalinis hyphis filimentosis et abundantia 6-14 µm lata cylindrica caulocystidia cum oleiferis extremis hyphis.

Holotypus: Lectus a R. M. Fatto 1115, sum quercu in Rutgers University's Hutcheson Memorial Forest, Somerset County, New Jersey, USA. 18 May 1998. Conservatus in herbario New York Botanical Garden (NY).

Pileus 3-9 cm broad, plano-convex with slightly inrolled margin, maturing plano-depressed; margin even, in age slightly striate to 5 mm; cuticle viscid, shiny, when dry remaining shiny, glabrous, separable to one-half the radius, disc area a deep neutral brown with hues about 58 moderate Brown, M&P 7J10, lighter marginally, 57 light Brown, M&P 15C10; trama thin, 4 mm at mid-radius, brittle, white, unchanging when injured, taste slowly moderately acrid, odor nil. **Lamellae** 6 mm broad at mid-radius, subacute in front, adnate to adnexed, close to medium, with rare long lamellulae or forking near the stipe, intervenose, edges even, brittle, white, unchanging. **Stipe** 2.5-6.5 x 0.7-2.5 cm, equal to slightly enlarging basally, dry, glabrous, longitudinally rugulose, white, stuffed, unchanging when injured.

Chemical reactions (stipe surface and trama): SV nil; 10% $FeSO_4$ pinkish-tan; 2% phenol brown; formalin nil; guaiac blue.

Spores white in mass, A or Ia, 8.0-10.0 x 6.0-7.2 µm, obovate, elliptic, broadly elliptic; ornamentation of amyloid cylindric to conic warts 0.5-1.0 µm high, isolated or with fine connectives occasionally frequent enough to form a broken reticulum; suprahilar area a weakly amyloid patch; hilar appendix inamyloid, conspicuous. **Basidia** 36-48 x 9-12 µm, clavate, 4-sterigmate. **Hymenial cystidia** 56-84 x 8-12 µm, fusoid-clavate, fusoid, lanceolate, apices acute, mucronate, infrequently with a short projection, filled or partially filled with refractive-grayish granules and short bands, incompletely SV+, arising from the subhymenium, projecting to 38 µm beyond the basidioles, abundant. **Subhymenium** 30-35 µm thick, upper portion pseudoparenchymatous, cells 6-10 µm broad, lower portion prosenchymatous, hyphae 2-4 µm wide. **Trama** with 3-6 µm wide,



FIGS. 10-13. *Russula primaverna* (Fatto 1115, holotype). 10. Spores. 11. Hymenial cystidia. 12. Hyphal ends from pileus cuticle. 13. Hyphal ends from stipe cuticle.

smooth vascular hyphae and oleiferous hyphae. **Pileus subcutis** 60-90 µm thick, of hyaline, horizontal-interwoven, filamentous hyphae 1.5-4 µm wide; also with smooth vascular and oleiferous hyphae. **Pileus epicutis** 130-200 µm thick, embedded in a gelatinous matrix, of interwoven, ascending to erect, hyaline, septate, 3-6 µm wide hyphae and abundant pileocystidia as ends of oleiferous hyphae, 3-8 µm wide, subcylindric, the terminal 50 µm usually strangulated, filled with refractive-grayish granular content, moderately SV+, aseptate or with a rare septum, apices rounded, occasionally subcapitate or with a finger like projection, originating in the epicutis, subcutis, or trama. **Stipe cuticle** a thin cutis of hyaline 2-4 µm wide hyphae and abundant caulocystidia as terminations of oleiferous hyphae, 40-140 x 6-14 µm, subcylindric, rather uniform in width, filled with refractive-grayish granular content, moderately SV+, apices rounded, aseptate or with a rare septum; also with uncommon smooth vascular hyphae.

Habitat scattered to solitary on soil under oak; May.

MATERIAL EXAMINED: NEW JERSEY: Somerset Co.: Rutgers University's Hutcheson Memorial Forest, 18 May 1998, Fatto 1115 (Holotype, NY). PENNSYLVANIA: Bucks Co.: Bowman's Hill Wildflower Preserve, 23 May 1998, Fatto 1116.

OBSERVATIONS: *Russula primaverna* is placed in subsection *Emeticinae* of section *Russula* as detailed by Shaffer (1975) which corresponds to Singer's subsection *Russula* of section *Russula*. Field characteristics aiding identification are the shiny, neutral brown pileus, white stipe and lamellae, unchanging trama, slowly moderately acrid taste, and deciduous, predominantly oak, habitat. The white, moderately large spores and prolific oleiferous hyphal system with subcylindric terminations in the pileus and stipe cuticles are confirming laboratory diagnostics. This taxon is very distinctive and yet has not previously been observed, probably because it may be rather rare and perhaps because of its unusual early fruiting period of mid May. Russulas in this central area of New Jersey are seldom seen before the latter part of June. The May of 1998 was not unusually warm nor wet. Both fruiting locations were reinspected biweekly till the beginning of June but no additional fruitings were observed.

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MYCOTAXON

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A NEW SPECIES OF ASCOTRICHA WITH UNIQUELY MOUNDED ASCOSPORES

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ABSTRACT

Ascotricha crucispora, a new species with a *Dicyma* anamorph, is described and illustrated from soil in the West Indies and Turkey. It is characterized by ostiolate ascomata with sparse geniculate hairs, cylindrical asci with an amyloid apex, and discoid ascospores which have longitudinal mounds on the convex surface. The *Dicyma* anamorph is characterized by verticillately arranged ampulliform conidiogenous cells on long conidiophores and nearly globose, smooth-walled, sympodioconidia. The holotype was isolated from coastal soil in Antigua and Barbuda.

Key words: *Ascotricha*, *Dicyma*, soil fungus, systematics.

In the course of exploratory survey of soil-borne ascomycetes as producers of secondary metabolites, we isolated two new species of *Ascotricha* Berk. (Xylariaceae): *A. distans* Udagawa, Uchiyama et Kamiya and *A. novae-caledoniae* Udagawa, Uchiyama et Kamiya (Udagawa et al., 1994a, b). A continuous survey recently revealed the isolation of a third unusual member of the genus from soil samples from the West Indies and Turkey. It is described below as a new species.

TAXONOMIC PART

Ascotricha crucispora Udagawa et Uchiyama, sp. nov. Figs. 1-9

Coloniae in agaro farinae avenaceae plus minusve celeriter crescentes, radiatim sulcatae, ex coacto mycelio tenui constantes, abundantibus ascomatibus formantes, viridi-griseae; conidiogenesis sparsa; reversum dilute aurantiacum.

Ascomata superficialia vel immersa, dispersa, ostiolata, atrobrunnea vel nigra, subglobosa vel ovoidea, 120-200×100-150 μm, cum collo brevi-conico instructa, paucipilosa; peridium brunneum vel atrobrunneum, tenue, ex "textura intricata" vel "textura epidermoidea" compositum; pili rigidi, sparsi, valde olivaceo-brunnei, vulgo geniculati, simplices vel pauci-ramosi, cum cellulis ampulliformibus (ramis) hyalinis instructi, 30-100 × 2-3 μm, incrassati, leves vel noduliferi, remote septati, ad apicem cellula ampulliformi hyalina formantes; cellulae ampulliformes 8-16×3-4 μm. Asci 8-spori, cylindracei, 60-82 × 12-16 μm, apparatus apicalis in mixtura Melzeri caerulescens, brevi-stipitati; paraphyses numerosae, hyalinae, filiformes. Ascosporae uniseriatae, primum hyalinae, deinde atrobrunneae, discoideae sed instar patellae volucris fictae, late ellipsoideae a fronte visis, fusiformes vel anguste ellipsoideae a latere visis, cruciformes a extremo visis, 12-14×9-11×5.5-8 μm, leves, incrassatae, fissura germinali longitudinaliter paratae. Status anamorphus: *Dicyma* sp.

Holotypus: SUM 3113, colonia exsiccata in cultura ex solo littoralibus, Antigua-Barbuda Insul., Antillae, 20.xii.1995, a S. Uchiyama isolata et ea collectione fungorum Musei et Instituti Historiae Naturalis Chiba (CBM) conservata.

Etymology : from Latin *cruci-* + *-sporus*, referring the shape of the ascospores in end view.

Anamorphosis: *Dicyma* sp.

Mycelium ex hyphis hyalinis vel dilute brunneis, ramosis, septatis, levibus compositum. Conidiophora erecta, ex mycelio basali oriunda; stipites 60-175×2-3 μm, leves vel noduliferi, incrassati, septati, olivaceo-brunnei, apicaliter verticillate ramosi; rami 2-5 verticillati, cylindracei, 12-55×2-3 μm. Cellulae conidiogenae hyalinae, cylindraceae vel ampulliformes, 5-20×3-5 μm, leves vel subtiliter asperatae, tenues, ad apicem denticulatae. Conidia hyalina, globosa vel late ovoidea, 3.5-5.5×3-4.5 μm, ad basim cicatrice inconspicua affixa, levia.

Holotypus: SUM 3113, loc. cit.

Colonies on oatmeal agar growing rather rapidly, attaining a diameter of 30-35 mm in 21 days at 25℃, radially sulcate, consisting of a thin mycelial felt and sparse aerial hyphae, granular in appearance due to the abundant production of black ascomata, Greenish Grey (M. 28F2, after Kornerup and

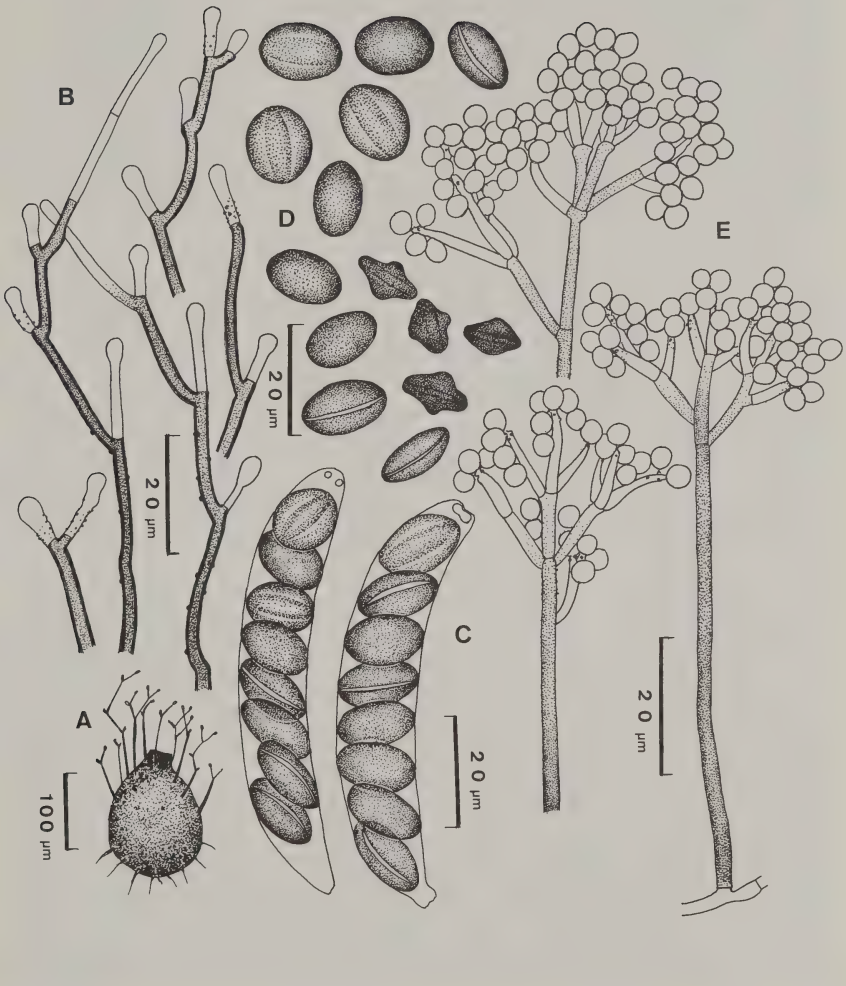
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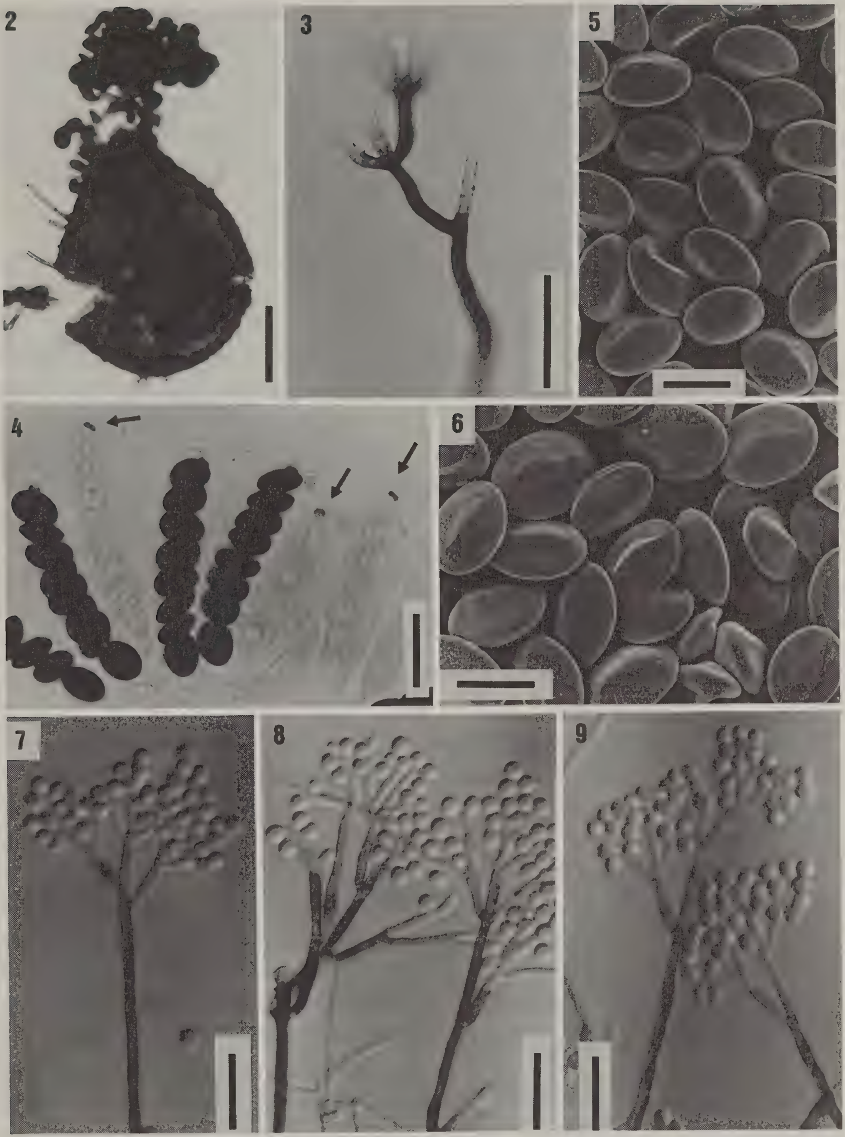
Fig. 1. *Ascotricha crucispora.*

A. Ascoma. B. Ascomatal hairs. C. Asci. D. Ascospores. E. Conidiophores and conidia.

Wanscher, 1978) or Mouse Grey (Rayner, 1970); margins thin and broad; conidiogenesis sparse, not influencing the colony appearance; exudate small, clear; reverse Pale Orange (M. 6A3) or Salmon (R). Colonies on potato-carrot agar growing slowly, attaining a diameter of 25-27 mm in 21 days at 25℃, somewhat floccose, plane, consisting of a thin mycelial felt, producing abundant ascomata on the felt, which are loosely covered by aerial hyphae and intermixed with profuse conidiophores; conidiogenesis moderate to conspicuous, Brownish Grey (M. 12F2) or Fuscous Black (R); reverse Pale Red (M. 7A3) or Flesh (R).

Ascomata superficial or sometimes immersed, scattered, ostiolate, dark brown to black, subglobose to ovoid, 120-200×100-150 μm, with a short conical neck at the apex, measuring 25-40 μm high and 40-45 μm wide; peridium brown to dark brown, thin, 8-12 μm thick, of "textura intricata" to "textura epidermoidea"; outer layer consisting of olive brown, thick-walled cells measuring 2-6 μm diam, and inner layer of pale brown, thin-walled, irregular cells. Ascomatal hairs terminal and lateral, rigid, few in number, scattered, dark olive brown, straight to mostly geniculate, simple or sometimes a few branched with hyaline ampulliform cells, 30-100×2-3 μm, thick-walled, smooth or nodulose, remotely septate, gradually tapering and paling above to a hyaline ampulliform cell; ampulliform cells 8-16×3-4 μm. Asci 8-spored, borne in a basal fascicle, cylindrical, 60-82×12-16 μm, rounded above, with apical apparatus bluing in Melzer's iodine reagent, discoid, 1.5-2.5 μm high and 3-4 μm wide, short-stipitate, thin-walled but rather persistent; paraphyses numerous, hyaline, filiform, 2-6 μm in diam, septate, mixed with the asci. Ascospores uniseriate, discharged through the ostiole as a black mass, at first hyaline, becoming dark brown, one-celled, discoid with longitudinal mounds, ellipsoidal flying-saucer in appearance, in face view broadly ellipsoidal with a fusoid-ellipsoidal raised area running lengthwise down the center, in side view fusiform to narrowly ellipsoidal, in end view cruciform, 12-14×9-11×5.5-8 μm, smooth, thick-walled, with a longitudinal germ slit running up to the entire length.

Mycelium consisting of hyaline to pale brown, branched, septate, smooth-walled hyphae. Conidiophores erect, mononematous, arising from the basal mycelium; stipes 60-175×2-3 μm, smooth or nodulose, thick-walled, septate, olive brown, paling and roughened above, apically often branched a few times in somewhat verticillate arrangements; branches in 2-5 verticils, cylindrical, 12-55×2-3 μm, ending in one to three conidiogenous cells or sometimes sterile ampulliform cells. Conidia hyaline, cylindrical to ampulliform, 5-20 × 3-5 μm, smooth or finely roughened, thin-walled, denticulate in clusters along the apical part of the rachis. Conidia hyaline, globose to broadly ovoid, 3.5-5.5×3-4.5 μm, with an inconspicuous basal



Figs. 2-9. *Ascotricha crucispora.*

2. Ascoma. 3. Ascomatal hair. 4. Asci in Melzer's reagent (arrows: amyloid apical apparatus). 5, 6. Ascospores (showing various views by SEM). 7-9. Conidiophores and conidia. Scale bars: Fig. 2 = 50 µm; Figs. 3, 4 = 20 µm; Figs. 5, 6 = 10 µm; and Figs. 7-9 = 20 µm.

scar, smooth-walled.

At 37℃, growth is nil.

Specimens examined: SUM 3113 (holotype), in dried culture isolated from coastal soil, Antigua and Barbuda, the West Indies, 20 October 1995, by S. Uchiyama; and SUM 3114, in culture isolated from Troy, Turkey, 28 September 1995. The holotype has been deposited with the Natural History Museum and Institute, Chiba, Japan (CBM).

NOTE. *Ascotricha crucispora* is a very distinctive fungus, and unlikely to be confused with any other so far known (Hawksworth, 1971; Kulshreshtha et al.,1977; Udagawa et al., 1994a, b; Stichigel and Guarro, 1998). The outstanding features of this species are: (1) ostiolate ascomata with sparse, geniculate hairs measuring 2-3 µm in diam, (2) cylindrical asci with a distinct amyloid apical apparatus, (3) large (12-14 µm long) discoid ascospores with longitudinal mounds which are seen as cruciform in end view, (4) somewhat verticillately arranged conidiogenous cells produced on long conidiophores, and (5) nearly globose, smooth-walled conidia. The amyloid asci and larger ascospores suggest an alliance with *A. erinacea* Zambett. (Khan and Cain, 1977), but the latter species differs in having abundantly branched, spinose terminal hairs, narrowly ellipsoidal ascospores (11-16 × 6.5-9 µm) and irregularly pyriform conidia. The geniculate terminal hairs and the total conidial picture are rather similar to those of *A. chartarum* Berk. (Ames, 1961; Hawksworth, 1971; Arx, 1982), but seems to be most easily distinguished by the ascospore shape and size; the ascospores of *A. chartarum* are flattened on two convex surfaces and tend to be smaller, measuring 7-9 × 7-8 µm.

Khan and Cain (loc. cit.) first found asci with an amyloid apical plug in *A. erinacea*, although most species of the genus produce asci characteristic of thin, evanescent walls and a non-amyloid apex (Roberts et al., 1984; Hanlin, 1990). The one-celled, dark ascospores containing an elongate germ slit and the holoblastic *Dicyma* anamorph are also supported to arrange *Ascotricha* in the Xylariaceae (Khan and Cain, 1977; Eriksson and Hawksworth, 1993; Hawksworth, in Barr and Cannon, 1994; Hawksworth et al., 1995). On the otherhand, Laessφe (1994) treated this genus as an exclusion from the Xylariaceae. He stated that the amyloid plug and type of anamorph seem to favor position within the Xylariaceae but e.g. the non-stromatic nature is highly unusual within that family. The formation of amyloid asci in *A. crucispora* further emphasizes the affinity of *Ascotricha* to the Xylariaceae, rather than in the Sordariales.

Ascotricha crucispora is also peculiar in its genus in the formation of flying-saucer-shaped ascospores but this character is hardly of particular significance above the species level, because some species of *Coniochaeta* share this same characteristic. Similar uniquely mounded ascospores have been unexpectedly encountered in *Coniochaeta extramundana* Mahoney et LaFavre and *C. cruciata* Fort et Guarro (Mahoney and LaFavre, 1981; Checa et al., 1988).

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CRYPTUMBELLATA, A NEW HYPHOMYCETE GENUS FROM SOIL

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ABSTRACT

Cryptumbellata terricola gen. et sp. nov. is described and illustrated together with its *Chrysosporium* synanamorph. Its sporodochia bear a superficial resemblance to conidial apparatus of *Oncocladium flavum*, but differ from them in having terminally trichotomously branched, uncinate appendages and an "aleurioconidial" mass. The fungus is also characterized by its ability to degrade keratin.

Key words: *Chrysosporium*, *Cryptumbellata terricola*, Hyphomycetes, soil fungus, systematics.

In an exploratory survey of soil-borne fungi as producers of secondary metabolites useful to the pharmaceutical industry, we recently isolated several new hyphomycetes from Japanese and overseas soils. One of them is an isolate producing appendaged sporodochia similar to *Oncocladium* Wallroth. After a thorough review of the literature (Orr and Kuehn, 1963, 1971; Apinis, 1964; Hughes, 1968; Sigler and Carmichael, 1976; Eckblad, 1981; Sigler et al., 1987), it was determined that this fungus represents a new genus and species which is described and illustrated herein.

TAXONOMIC PART

Cryptumbellata Udagawa et Uchiyama, gen. nov.

Genus Hyphomycetum.

Coloniae modice crescentes, floccosae vel funiculosae, albae vel dilute brunneo-aurantiacae. Mycelium ex hyphis hyalinis, ramosis, septatis, levibus compositum. Sporodochia superficialia, dispersa, sessilia, dilute brunnea, sphaerica, ex massa conidiorum holoblastico et appendices hypharum circumcinctis composita; appendices primum ex mycelio juveni radiatim oriundae, stipites principales erecti, firmi, dilute flavo-brunnei vel hyalini, crassitunicati, leves, septati, inferne in ramos laterales fertiles efferentes, superne ramosi, ad medium ramulis duobus sinuolatis lateraliter formantes, ad extremum ramulis trichotomis uncinatis formantes. Stromata nulla. Conidiophora saepe indistincta, ex stipitibus appendicum basaliter et lateraliter exorientia, hyalina, tenuitunicata, levia, septata, irregulariter ramosa. Cellulae conidiogenae monoblasticae vel polyblasticae, discretae, terminales vel laterales, hyalinae, brevi-cylindraceae. Conidia holoblastica, singula, simplicia, sicca, unicellularia, subhyalina, subglobosa vel obovoidea, basi truncata, crassitunicata, verruculosa.

Species typica: *Cryptumbellata terricola* Udagawa et Uchiyama.

Synanamorphosis: *Chrysosporium* Corda.

Etymology: Latin, *crypt-* = covered and *umbellata* = furnished with partial umbels, referring to the characteristics of the appendaged sporodochia.

A genus of Hyphomycetes. Colonies moderately growing, floccose to funiculose, white to pale brownish orange. Mycelium composed of hyaline, branched, septate, smooth-walled hyphae. Sporodochia superficial, scattered, sessile, pale brown, spherical, consisting of a mass of holoblastic conidia surrounded by hyphal appendages; appendages at first arising radially from the young mycelium, consisting of main stalks which are erect, firm, pale yellowish brown to hyaline, thick-walled, smooth and septate, in the lower part of the stalk producing lateral fertile branches, in the upper part branched, forming two, faintly sinuous, lateral branchlets at the middle node and trichotomous, uncinately incurved branchlets at the terminal. Stromata none. Conidiophores often indistinct, arising basally and laterally from the stalks of appendages, hyaline, thin-walled, smooth, septate, irregularly branched. Conidiogenous cells monoblastic and polyblastic, discrete, terminally or laterally borne, hyaline, short-cylindrical. Conidia holoblastic, solitary, simple, dry, one-celled, subhyaline, subglobose to obovoid, basally truncated, thick-walled, verruculose.

Type species: *Cryptumbellata terricola* Udagawa et Uchiyama.

Synanamorph: *Chrysosporium* Corda.

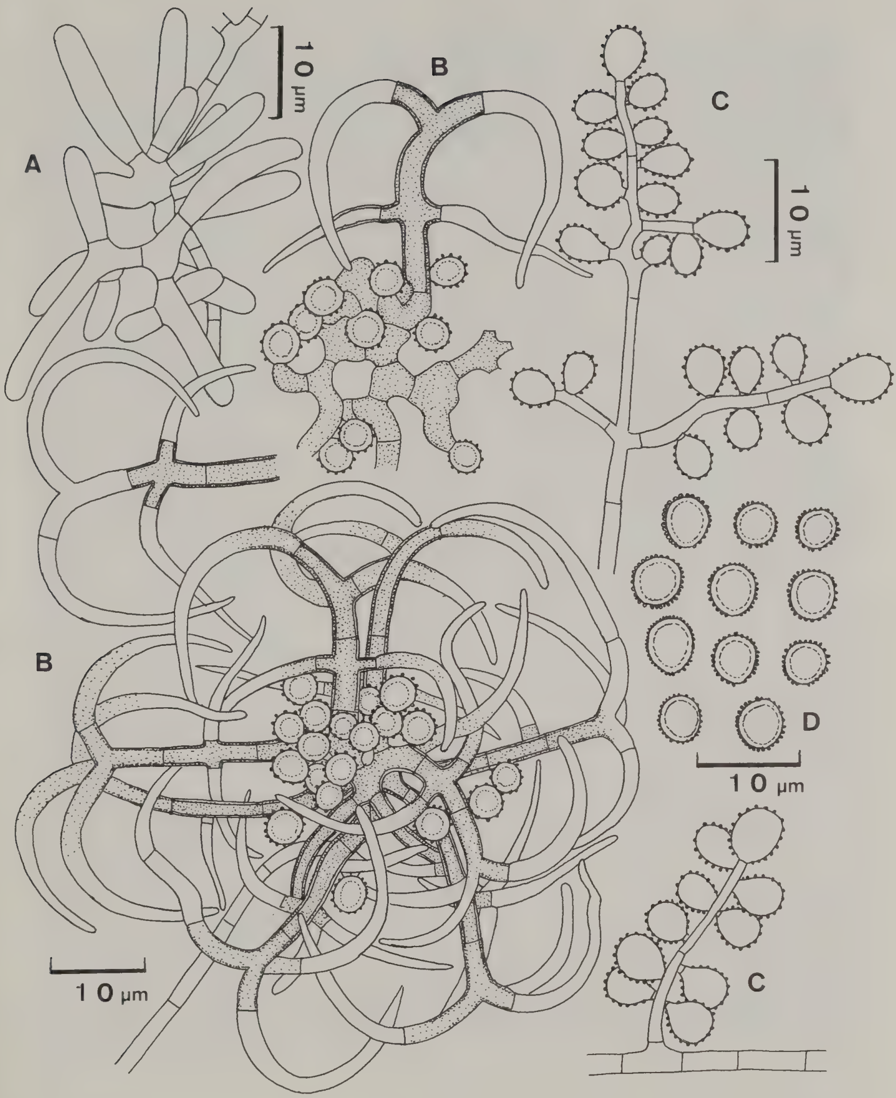
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Fig. 1. *Cryptumbellata terricola.*

A. Initial development of sporodochium. B. Sporodochia, showing hyphal appendages, conidiogenous cells and conidia. C, D. Conidiophores and conidia of the *Chrysosporium* synanamorph.

Cryptumbellata terricola Udagawa et Uchiyama, sp. nov. Figs. 1-11

Coloniae in agaro cum decocto tuberorum et carotarum modice crescentes, floccosae vel funiculosae, planae, ex mycelio basali coacto tenuiter constantes, albae vel dilute brunneo-aurantiacae; conidiogenesis abundus; reversum griseo-brunneum.

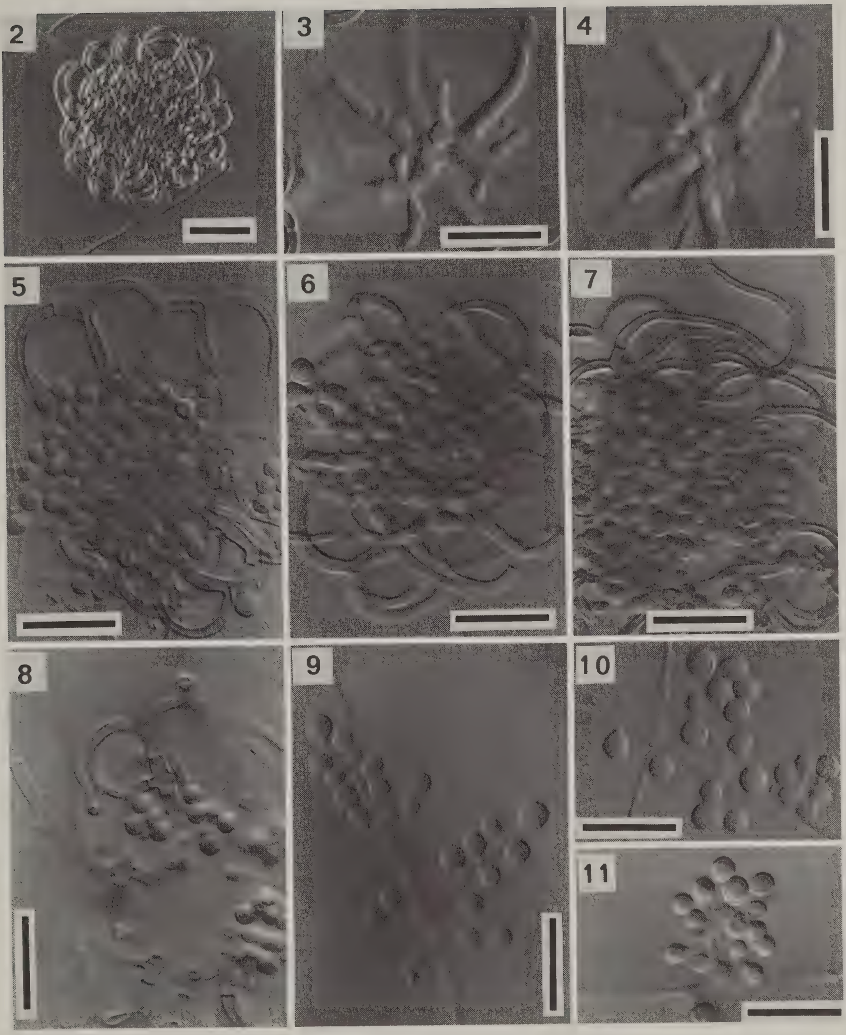
Mycelium ex hyphis hyalinis, ramosis, septatis, levibus, 1-2.5 µm diam compositum. Sporodochia dilute flavo-brunnea vel dilute brunnea, sphaerica vel subsphaerica, 60-100 µm diam cum appendicibus peripheralibus; appendices ex mycelio juveni radiatim oriundae, stipites principales erecti, firmi, dilute flavo-brunnei vel hyalini, crassitunicati, leves, septati, 20-30 × 2.5-4 µm, inferne in ramos laterales fertiles efferentes, superne ramosi, ad medium ramulis duobus sinuolatis formantes, ad extremum ramulis trichotomis uncinatis formantes. Conidiophora saepe indistincta, stipitibus appendicum basaliter et lateraliter exorientia, hyalina, tenuitunicata, levia, septata, irregulariter ramosa. Cellulae conidiogenae monoblasticae vel polyblasticae, discretae, terminales vel laterales, hyalinae, brevi-cylindraceae, 4-7 × 1.5-3 µm. Conidia subhyalina, subglobosa vel obovoidea, 2.5-6 × 2.5-5 µm, basi truncata, crassitunicata, verruculosa.

Chrysosporium synanamorphosis. Conidiophora solitaria, sparsim ramosa, saepe indistincta, hyalina, 20-100 × 1.5-2.5 µm, tenuitunicata, levia, septata. Conidia terminalia et lateralia numerosa, sessilia vel ex pedicellis brevibus oriunda, solitaria, subhyalina, subglobosa, obovoidea vel pyriformia, 3-5.5(-7) × 2.5-4.5(-6.5) µm, basi truncata, tenuitunicata, verruculosa. Conidia intercalaria minus numerosa, subhyalina, levia, tenuitunicata, cylindracea vel doliiformia, 1-2-cellularia, 4-12.5 × 1.5-3 µm, utrinque truncata. Chlamydosporae absentes. Teleomorphosis abest.

Holotypus. SUM 3115: colonia exsiccata in cultura ex solo prope alveum espulae, prope Menghai, Yunnan in Sina, 7.viii.1994, a S. Uchiyama isolata et ea collectione fungorum Musei et Instituti Historiae Naturalis Chiba (CBM) conservata.

Etymology: Latin, *terricola* = dwelling on the ground, referring to its habitat.

Colonies on potato-carrot agar (PCA) growing moderately, attaining a diam of 34-37 mm in 14 d at 25°C, floccose to funiculose, plane, consisting of a thin basal felt, White to Brownish Orange (M. 6C3, after Kornerup and Wanscher, 1978) or Vinaceous Buff to Hazel (Rayner, 1970), producing abundant sporodochia with an admixture of loose aerial hyphae and solitary conidiophores; margins entire, thin; exudate small, clear; odor suggestive of mushrooms; reverse Greyish Brown (M. 6D3) or somewhat Greyish Sepia (R).



Figs. 2-11. *Cryptumbellata terricola.*

2. Sporodochium. 3, 4. Initial development of sporodochia. 5-8. Magnification of mature sporodochia, showing hyphal appendages, conidiogenous cells and conidia. 9-11. Conidiophores and conidia of the *Chrysosporium* synanamorph. All scale bars = 20 μm.

Colonies on oatmeal agar growing moderately but somewhat more rapidly than on PCA, about 42-44 mm in 14 d at 25°C, more or less floccose, vegetative mycelium thin and submerged, Greyish Brown (M. 7D3) or Vinaceous Buff (R); conidiogenesis abundant; reverse Light Brown (M. 6D4) or Fawn (R).

Mycelium composed of hyaline, branched, septate, smooth-walled, 1-2.5 µm diam hyphae; racquet hyphae absent. Sporodochia pale yellowish brown to pale brown, spherical to subspherical, 60-100 µm in diam incl. peripheral appendages; appendages arising as radially branching clusters from the young mycelium, consisting of a thick-walled main stalk and thin-walled, swollen side branches which are irregularly growing out into a compact whorl of conidiogenous cells in the lower part; the main stalks erect, firm, pale yellowish brown and fading toward the hyaline apex, smooth-walled, septate, 20-30 × 2.5-4 µm, branched above the fertile region, forming two lateral branchlets at the middle node and trichotomous, uncinately incurved, up to 25-35 µm long branchlets at the terminal. Conidiophores often indistinct, short, borne as the side branches on the basal portion of appendage stalk more or less at right angles, hyaline, thin and smooth-walled, septate, irregularly rebranched. Conidiogenous cells monoblastic and polyblastic, discrete, produced terminally and laterally, hyaline, short-cylindrical, 4-7 × 1.5-3 µm. Conidia subhyaline, subglobose to obovoid, 2.5-6 × 2.5-5 µm, basally truncated, thick-walled, verruculose.

Chrysosporium synanamorph. Conidiophores solitary, sparsely branched, often undifferentiated, hyaline, 20-100 × 1.5-2.5 µm, thin-walled, septate. Conidia mostly terminal or lateral, sessile or on short pedicels, solitary, subhyaline, subglobose, obovoid or pyriform, 3-5.5 (-7) × 2.5-4.5 (-6.5) µm, basally truncated, with walls thin and verruculose; intercalary conidia subhyaline, smooth and thin-walled, cylindrical or barrel-shaped, 1-2-celled, 4-12.5 × 1.5-3 µm, truncated at both ends. Chlamydospores absent.

At 37°C, growth is nil.

Keratinolytic (tested for ability to digest keratin by placing a number of cut fine horse hairs on the water agar 5 mm away from the point of inoculation).

Holotype. SUM 3115: dried culture derived from an isolate from soil associated with beehive of *Vespula*, near Menghai, Yunnan, China, 7 August 1994, collected by S. Uchiyama. The type specimen is deposited in the Natural History Museum and Institute, Chiba, Japan (CBM).

NOTE. The outstanding features of *Cryptumbellata terricola* are: (1) pale brown, spherical sporodochia which develop as a cluster of radially branching hyphae on the basal mycelium and composed of thick-walled, terminally trichotomously branched, uncinate appendages and a central mass of "aleurioconidia," (2) conidiophores arising from the main stalk of hyphal appendages as lateral branches in the basal portion; (3) monoblastic and polyblastic, discrete, short-cylindrical conidiogenous cells, (4) subhyaline, one-celled, subglobose to obovoid, basally truncated, verruculose conidia, and (5) keratinophilic growth. The *Chrysosporium* synanamorph is somewhat similar to *C. carmichaelii* van Oorschot (van Oorschot, 1980), but differs by terminal and lateral conidia which tend to be subglobose rather than clavate and its ability to utilize keratin.

The genus *Cryptumbellata* and its *Chrysosporium* synanamorph are reminiscent of, but distinct from, *Gymnoascus*-like pleoanamorphic fungus *Oncocladium* described and illustrated for *O. flavum* Wallroth by Hughes (1968) and Sigler et al. (1987). *Oncocladium flavum* is a striking fungus characterized by pale yellow brown, thick-walled hyphal appendages which bear regular verticils of 3-5 reflexed branchlets. The branchlets in the basal region are less curved, bearing hyaline hyphal filaments which divide to form alternate arthroconidia and undergo rhexolytic dehiscence (Sigler et al., 1987). The verticillately-branched hyphae of *O. flavum* have not been obtained in agar culture, and were known only from keratinous substrates in ascoma-like conidial apparatus which resemble the mesh-like gymnothecia typical of several members of the Onygenales. Closely associated with the verticillately-branched hyphae is an arthroconidial anamorph referred to *Malbranchea*, the synanmorph, which has been easily cultured (Sigler and Carmichael, 1976).

The hyphal appendages of *C. terricola* are readily distinguished by the presence of terminally trichotomous branches with an uncinate end. In contrast to the arthroconidia of *O. flavum* and its *Malbranchea* synanamorph, conidiogenesis of the both morphs of *C. terricola* is characterized by holoblastic development.

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CULTURAL CHARACTERISTICS AND TAXONOMY OF SOME POLYPORES (APHYLLOPHORALES) FROM SANTA CATARINA ISLAND, SC, BRAZIL

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ABSTRACT

Cultural characters of seven wood-rotting polypores (Aphyllophorales) collected in Santa Catarina Island (Southern Brazil) were studied according to Nobles' system (1965). Little or nothing was known of the cultural characteristics of these polypores. The species are *Phellinus apiahynus* (Speg.) Rajchenb. & Wright, *Antrodiella multipileata* Loguercio-Leite & Wright, *Fomitella supina* (Sw.: Fr.) Murr., *Megasporoporia cavernulosa* (Berk.) Ryv., *Perenniporia stipitata* Ryv., *Rigidoporus microporus* (Fr.) Overeem and *Trametes pavonia* (Hook.) Ryv. The cultural characteristics were helpful in distinguishing the species, specially in *Fomitella supina*, *Antrodiella multipileata* and *Trametes pavonia*.

KEY WORDS: cultural studies, Aphyllophorales, wood rotting

INTRODUCTION

Santa Catarina Island (27° 10' and 27° 30' S lat. and 48° 25' and 48° 35' W long.) is a subtropical area, whose vegetation corresponds to the Atlantic Rain Forest in a secondary stage (IBGE, 1992). During the last ten years collecting of wood-rotting fungi has resulted in new records of polypores to Santa Catarina (Loguercio-Leite & Wright, 1991a, 1991b; Gerber & Loguercio-Leite, 1997, Loguercio-Leite & Gerber, 1997).

In this paper cultural studies are reported for 7 polypores, in order to better understand their biology, and consequently, their taxonomy. The cultural features of five species were unknown. Six of the species are well known by mycologists, while *A. multipileata* has been reported only once (Loguercio-Leite & Wright, 1991b).

MATERIAL AND METHODS

Cultures were isolated from tissues of fresh fruitbodies collected on angiosperms in Santa Catarina Island. They were studied and described according to Nobles (1965). The number after 'cc' indicates the culture while the number after FLOR indicates the voucher specimen.

Dicaryotic cultures were inoculated in the center of 10 cm Petri dishes with malt extract agar (MEA), differing form Nobles (1965) which inoculated the dishes at the side. For this reason the code symbol to rate of growth was not included. Cultures were grown on MEA in the dark at 25° C and studied during 6 weeks. Microscopic preparations were mounted in KOH 5% plus phloxine 1% and in Melzer's reagent. Drawings were made with a camera lucida (1000X).

Tests on gallic acid (GAA) and on tannic acid (TAA) agar media were recorded at 1 week after inoculation at the same conditions (dark, 25° C) and were made according to Nobles (1965) while the intensity was evaluated based on Bettucci and Guerrero (1971). Biochemical enzymatic tests were performed according to Käärik (1965), Harkin and Obst (1973), Taylor (1974) and Stalpers (1978). The cultures were 7 days old when the biochemical tests were performed. The intensity of reactions are classified in: negative (-), weak (+), medium (++) and strong (+++).

RESULTS AND DISCUSSION

This work allows to improve the knowledgement from the mycelian characteristics of the species described here in culture and increases the number of characters that can be used in traditional taxonomy. Species are in two families, one of them belongs to the Hymenochaetaceae (*Phellinus apiahynus*) while the six others belong to the Polyporaceae (*Antrodiella multipileata*, *Fomitella supina*, *Megasporoporia cavernulosa*, *Perenniporia stipitata*, *Rigidoporus microporus* and *Trametes pavonia*).

Phellinus apiahynus (Speg.) Rajchenb. & Wright Fig. 1
Mycologia, 79 (2): 251. 1987.

Cultures studied: cc 470 (≅ FLOR 11000; BRAZIL, SC, Ilha de Santa Catarina, Morro da Lagoa; Gerber & Cabral; 30/V/1994).

Macroscopic characters: Mat cottony, yellowish brown to pale yellow at the margin; margin regular; growing very slowly, 7,5 - 8,0 cm diam. at the 6th week. Reverse darkened. Odor faint.

Microscopic characters: Marginal hyphae simple-septate, thin-walled, with some small ramifications (1,5 - 4,0 μm diam.). After three weeks the number of ramifications decrease. Fiber hyphae unbranched, hyaline, thick walled, mostly present at the center of the culture (2,0 - 4,0 μm diam.), sometimes presenting small projections. Sexual structures and setae absent.

Enzymatic tests: GAA: ++++; TAA: ++++; Laccase: ++(α-naphtol), -(guaiacol), ++(syringaldazine); Cytochromo oxidase: -(tmρda); Peroxidase: +(pyrogallol); Tyrosinase: -(ρ-cresol)

Species code: 2. 6. 8. 32. 37. 39. 50. 54.

Remarks: The cultural features of *P. apiahynus* are quite similar to those of *P. robustus* (Karst.) Bourd. & Galz., a species which basidiome is macroscopically similar to *P. apiahynus*. Stalpers' (1978) description of cultures of *P. robustus* differed from ours only in the formation of fewer fiber hyphae. The former species is distinguished by the small triangular basidiomes, and smaller spores (Rajchenberg and Wright, 1987). We could ask if there is real difference between these two species.

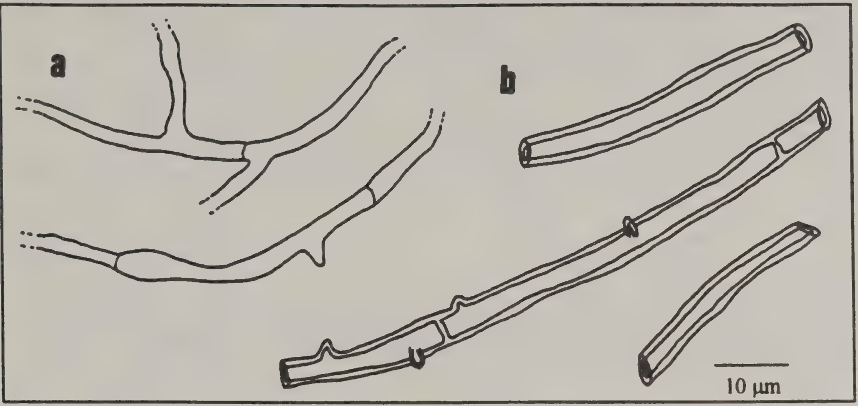

Fig. 1: *Phellinus apiahynus*. a) simple-septate hyphae; b) fiber hyphae

Antrodiella multipileata Loguercio-Leite & Wright — Fig. 2
Mycotaxon, 41 (1): 167. 1991.

Culture studied: cc 746 (≅ FLOR 11284; BRAZIL, SC, Ilha de Santa Catarina, Lagoa do Peri; Althoff, Gerber & Neves; 26/VII/1995).

Macroscopic characters: Mat cottony becoming felty in the center; white to pale yellow; margin regular; growing rapidly, dish covered by the 2nd week. Reverse unchanged. Odor strong, unpleasant.

Microscopic characters: Mycelium formed by clamped generative hyphae (3,0 - 6,0 μm diam.), aerial mycelium composed of fiber hyphae, with thin walls, not observed near the agar. After the 3rd week, branched binding hyphae develop in the aerial mycelium. After the 4th week some hyphae becoming wider forming a pseudoparenchymatous layer by the margin of the culture. Sexual structures absent.

Enzymatic tests: GAA: ++; TAA: ++++; Laccase: +(α-naphtol), -(guaiacol), ++(syringaldazine); Cytochromo oxidase: +(tmρda); Peroxidase: +(pyrogallol); Tyrosinase: -(ρ-cresol)

Species code: 2. 3. 8. 10. 32. 36. 38. 53. 54.

Remarks: *A. multipileta* presents an effused-reflexed to multipileate basidiomata with an irpicioid hymenial configuration, and broadly ellipsoid spores (3.6-5.1 μm long) (Loguercio-Leite and Wright, 1991b). In culture *A. multipileata* is trimitic as are *A. semisupina* (Berk. & Curt.) Ryv. and *A. citrinella* Niem. & Ryv. studied by David & Tortíc (1986). Microscopically, *A. semisupina* and *A. citrinella* are distinguished mainly by the spores size (2.5-3.5 μm long and 3-3.5 μm long, respectively). In these three species the generative hyphae differentiate into binding and skeletal hyphae and cultures produce laccase and peroxidase. In culture *A. multipileata* differs from the other two species in forming a pseudoparenchyma similar to figures 52 - 55 in Nobles (1965).

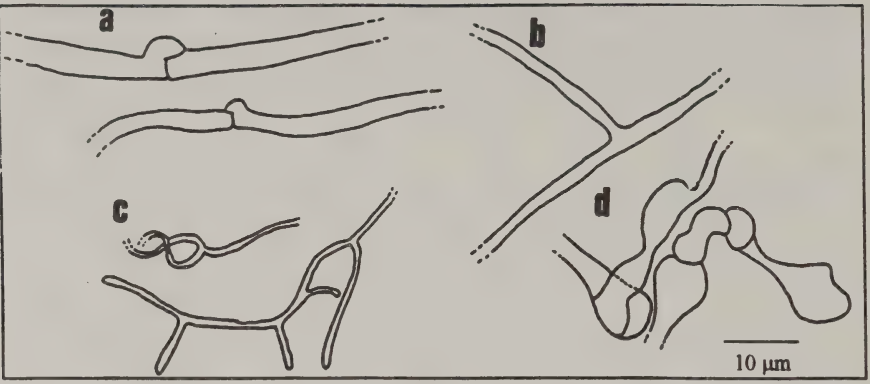

Fig. 2: *Antrodiella multipileata*. a) clamped generative hyphae; b) fiber hyphae; c) binding hyphae; d) pseudoparenchyma

Fomitella supina (Sw.: Fr.) Murr. Fig. 3
Bull. Torrey Bot. Club, 32: 365. 1905.

Cultures studied: cc 549 (≅ FLOR 11095; BRAZIL, SC, Ilha de Santa Catarina, Morro da Lagoa; Degenhardt, Halmenschlager & Neves; 22/IX/1994); cc 663 (≅ FLOR 11213; BRAZIL, SC, Ilha de Santa Catarina, Morro da Lagoa; Althoff, Gerber & Foresti; 30/V/1995).

Macroscopic characters: Mat cottony and white, becoming crustose and pale brown after the 3rd week; margin regular; growing rapidly, dish covered by the 2nd week. Reverse unchanged. Odor sweet.

Microscopic characters: Mycelium formed by generative clamped hyphae (3,0 - 6,0 µm diam.) thin and thick walled. Fiber hyphae thick walled (3,0 - 5,0 µm diam.) in the center of the colony and, after the 3rd week, also in the margin. Sexual structures absent.

Enzymatic tests: GAA: +++; TAA: ++++; Laccase: +++(α-naphtol), -(guaiacol), +++(syringaldazine); Cytochromo oxidase: +++(tmρda); Peroxidase: +++(pyrogallol); Tyrosinase: -(ρ-cresol)

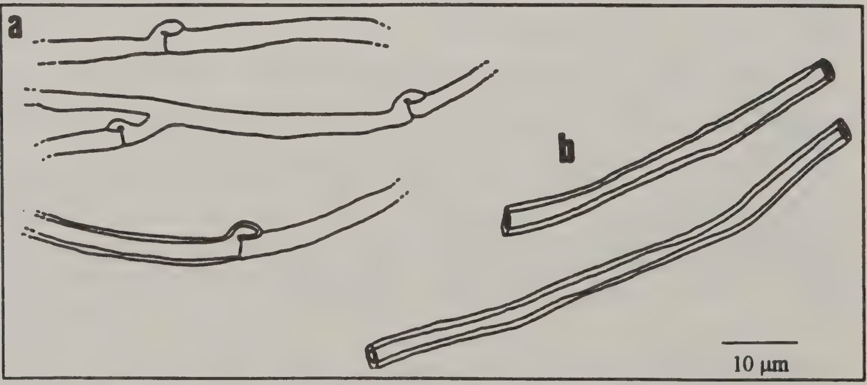

Fig. 3: *Fomitella supina*. a) clamped generative hyphae; b) fiber hyphae

Species code: 2. 3. 8. 32. 36. 38. 50. 54.
Remarks: *Fomitella supina* is the only species in this genus and it is morphologically similar with species of *Fomitopsis*. In fact, the character that distinguishes them is the type of rot (Gilbertson & Ryvarden, 1986), which in culture is seen as positive reaction in the enzimatic tests (GAA, TAA) for *Fomitella*. Cultures of some *Fomitopsis* species studied by Stalpers (1978) are very similar to our culture of *Fomitella supina*. Generally, *Fomitopsis* species produce chlamydospores in culture, but our cultures of *Fomitella supina* did not, thus this structure may be another character to distinguish these species, besides the type of rot.

Megasporoporia cavernulosa (Berk.) Ryv. Fig. 4
Mycotaxon, 16: 174. 1982.

Cultures studied: cc 575 (≅ FLOR 11083; BRAZIL, SC, Ilha de Santa Catarina, Morro da Lagoa; Degenhardt & Halmenschlager; 17/X/1994); cc 579 (≅ FLOR 11096; BRAZIL, SC, Ilha de Santa Catarina, Morro da Lagoa; Degenhardt & Halmenschlager; 17/X/1994); cc 608 (≅ FLOR 11341; BRAZIL, SC, Ilha de Santa Catarina, Morro da Lagoa; Althoff, Gerber & Foresti; 16/III/1995).
Macroscopic characters: Colony crustose and appressed in the center, cottony and raised in the margin, white to pale yellow, margin regular; growing slowly, dish covered by the 3rd week. Reverse unchanged. Odor musty.
Microscopic characters: Mycelium formed by clamped generative hyphae (3,0 - 8,0 µm diam.) that differentiate to dextrinoid fiber hyphae (2,0 - 5,0 µm diam.) after the 2nd week. At the 3rd week the fiber hyphae are predominant in the culture, generative hyphae being present only in aerial mycelium. Some few crystals are formed near the agar. Sexual structures absent.
Enzymatic tests: GAA: +++; TAA: +++; Laccase: ++(α-naphtol), -(guaiacol), +(syringaldazine); Cytochromo oxidase: -(tmρda); Peroxidase: +(pyrogallol); Tyrosinase: -(ρ-cresol)
Species code: 2. 3. 8. 32. 36. 38. 51. 54.

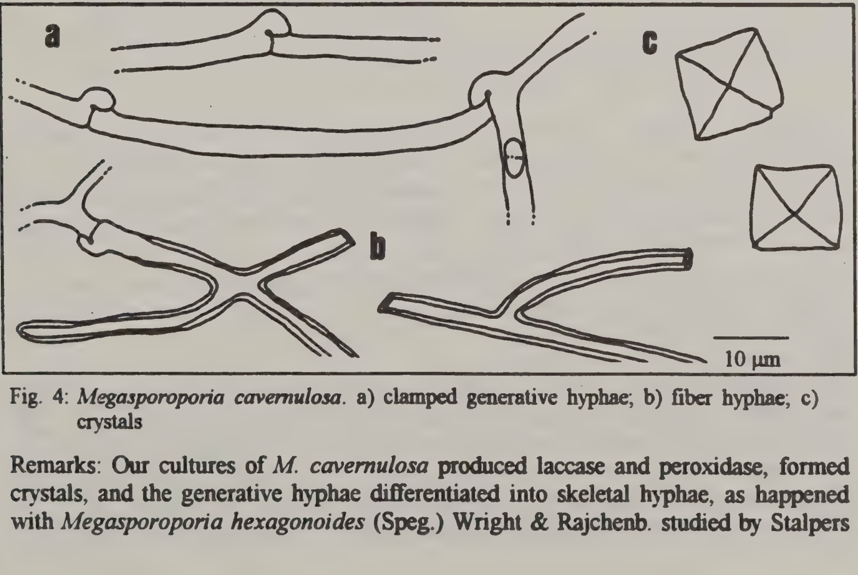

Fig. 4: *Megasporoporia cavernulosa*. a) clamped generative hyphae; b) fiber hyphae; c) crystals

Remarks: Our cultures of *M. cavernulosa* produced laccase and peroxidase, formed crystals, and the generative hyphae differentiated into skeletal hyphae, as happened with *Megasporoporia hexagonoides* (Speg.) Wright & Rajchenb. studied by Stalpers

(1978). These species are distinguished in their badidiomes by the presence or not of dendrohyphidia along the dissepiments and by the spores size. According to Ryvarden (1982) *M. cavernulosa* presents dendrohyphidia and has smaller spores (10,0-16,0 (18,0) µm) while *M. hexagonoides* does not present dendrohyphidia and the spores are longer (16,6-21,8 µm). It is important to emphasize that *M. cavernulosa* maintains a conspicuous dextrinoid reaction in skeletal hyphae, which is a distinctive characteristic of this genus. This reaction is lacking in cultures of *M. hexagonoides*, and could be useful character to distinguish them.

Perenniporia stipitata Ryv. Fig. 5
Mycotaxon, 28 (2): 535. 1987.

Culture studied: cc 499 (≅ FLOR 11015; BRAZIL, SC, Ilha de Santa Catarina, Morro da Lagoa; Cabral, Gerber & Halmenschlager; 28/VI/1994).
Macroscopic characters: Mat crustose and dense, white to the center, hyaline to the margin. After the 4th week the mat formed crustose elevations. Margin regular; growth very slow, 4,0 - 4,5 cm diam. at 6th week. Reverse unchanged. Odor faint.

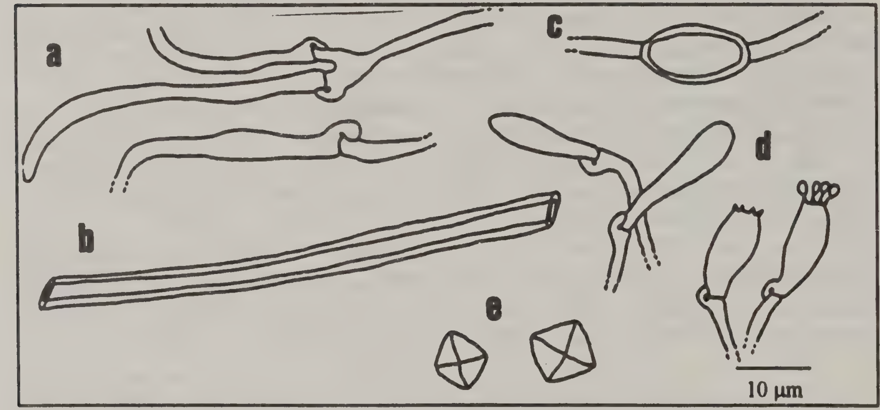

Fig. 5: *Perenniporia stipitata*. a) clamped generative hyphae; b) fiber hypha; c) chlamydospore; d) basidioles and basidia; e) crystals

Microscopic characters: Aerial and submerged mycelium formed by clamped generative hyphae (3,0 - 6,0 µm diam.). Fiber dextrinoid thick-walled hyphae (3,0 - 7,0 µm diam.) develop in the center submerged mycelium. Ellipsoid intercalary chlamydospores (17,0 x 25,0 µm diam.) present in crustose areas. Basidioles, basidia with sterigmata and, sometimes, with young basidiospores (1,0 µm) formed in the crustose areas after the 4th week. Marginal pyramidal crystals are present near the agar (12,0 - 15,0 x 12,0 - 15,0 µm diam.).
Enzymatic tests: GAA: +; TAA: +
Species code: 2. 3. 8. 34. 36. 38. 50. 54.
Remarks: Our culture of *P. stipitata* had a slow growth, not covering the plate within six weeks, produced skeletal dextrinoid hyphae and chlamydospores, all characteristics observed also to *P. martius* (Berk.) Ryv. studied by Wright and Deschamps (1975). The difference is that culture of *P. martius* produced binding hyphae. When we compared these cultures with a culture of *P. medulla-panis* (Fr.) Donk., also studied by Wright and Deschamps (*op cit*) we note that the last species

had faster growth and did not produce sexual structures. These three species can be separated by the following features of their fruitbodies: the first (*P. stipitata*) has a stipitate basidiomes, while the second (*P. martius*) is dimidiate, ungulate to lightly effused-reflexed, and the third (*P. medulla-panis*) is usually resupinate, becoming widely effused. *P. stipitata* was the only that produced basidioles and basidia in culture within six weeks.

Rigidoporus microporus (Fr.) Overeem Fig. 6
Icon. Fung. Malayensium, 5: 1. 1924.

Culture studied: cc 797 (≅ FLOR 11353; BRAZIL, SC, Ilha de Santa Catarina, Canto dos Araçás; Althoff, Gerber & Neves; 22/XI/1995).
Macroscopic characters: Mat hyaline, apressed without differentiation, excepting few little cottony tufts by the dish margin. Margin irregular; growth very rapid, dish covered by the first week. Reverse unchanged. Odor musty.
Microscopic characters: Mycelium formed by generative, simple-septate, thin or thick-walled hyphae (1,5 - 6,0 µm diam.). Fiber hyphae absent. Pyramidal (3,0 - 5,5 µm diam.) or irregular crystals present in the center of the mat, the last ones free or incrusted on generative hyphae, seen after the 3rd week. Cottony tufts at the margin composed of generative, simple-septate hyphae and few hyphae with clamps on a few of the septa (1,5 - 4,5 µm) seen only at last week. Sexual structures absent.
Enzymatic tests: GAA: +++; TAA: +++; Laccase: ++(α-naphtol), -(guaiacol), +++(syringaldazine); Cytochromo oxidase: +(tmρda); Peroxidase: ++(pyrogallol); Tyrosinase: -(ρ-cresol)
Species code: 2. 5. 7. 32. 36. 38. 51. 54.

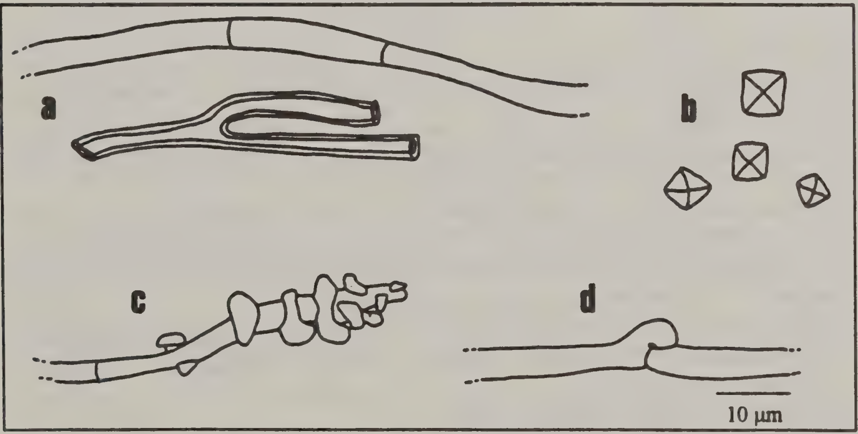

Fig. 6: *Rigidoporus microporus*. a) simple-septate hyphae; b) crystals; c) hyphae incrusted with crystals; d) clamped generative hypha

Remarks: This genus is characterized in the carpophore by simple-septate hyphae. *R. microporus* does not have cystidia and spore size is 3.5 - 5.0 µm long and differs from *R. lineatus* (Pers.) Ryv. that has cystidia and bigger spores (4.5 - 6.0µm long), both species are pileate. *R. vinctus* (Berk.) Ryv. is resupinate, does not have cystidia and the spores has an intermediate size (4.0 - 5.5µm long). *R. microporus* was studied in culture before by Bakshi *et al.* (1963) and Wright and Deschamps (1975) without the mention of clamped hyphae. In our study, a conspicuous characteristic in the culture was the presence of simple-septate and clamped hyphae together. Studies with *R. lineatus* (Bakshi *et al.*, *op. cit.*; Neves, 1996) and with *R. vinctus* (Hood & Dick, 1988) showed clamped hyphae in culture. Thus, clamped hyphae may be a useful taxonomic feature in *Rigidoporus* cultures.

Trametes pavonia (Hook.) Ryv. Fig. 7
Norw. J. Bot., 19: 237. 1972.

Culture studied: cc 545 (≅ FLOR 11088; BRAZIL, SC, Ilha de Santa Catarina, Morro da Lagoa; Degenhardt, Halmenschlager & Neves; 22/IX/1994); cc 601 (≅ FLOR 11063; BRAZIL, SC, Ilha de Santa Catarina, Morro da Lagoa; Althoff, Foresti & Gerber; 16/III/1995).

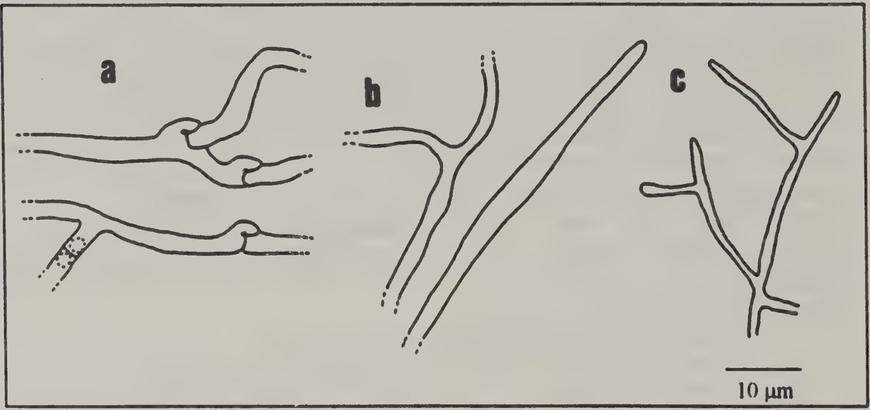

Fig. 7: *Trametes pavonia*. a) clamped generative hyphae; b) fiber hyphae; c) binding hyphae

Macroscopic characters: Mat cottony by the margin, crustose by the center and white. Margin regular; growth slow, dish covered by the 3rd week. Reverse unchanged. Odor musty.

Microscopic characters: Mycelium formed by generative clamped hyphae (3,5 - 7,0 (10,0) µm diam.). Fiber hyphae (2,0 - 5,0 µm diam.) formed after the 2nd week, most of them present in the center of the culture. Submerged binding hyphae present in the margin (0,5 - 1,0 µm diam.). Sexual structures absent.

Enzymatic tests: GAA: ++++; TAA: +++; Laccase: ++(α-naphtol), -(guaiacol), ++(syringaldazine); Cytochromo oxidase: -(tmρda); Peroxidase: -(pyrogallol); Tyrosinase: -(ρ-cresol)

Species code: 2. 3. 8. 32. 36. 38. 51. 54.

Remarks: *T. pavonia* has been confused with *Trametes villosa* (Fr.) Kreisel. In the former the basidiocarp is flabeliform to dimidiate, zonate, hirsute to glabrous while the second has a hirsute, thin and pliable basidiocarp. In culture *T. pavonia* has a trimitic hyphal system. Another culture of this species studied by Rajchenberg (1982) had the same results. Small differences include swellings on hyphae and the reverse bleached in the culture studied by Rajchenberg (*op. cit.*). Bettucci and Guerrero (1971) studied *T. villosa* in culture describing faster growing mycelium, smaller diameter generative and fiber hyphae, and presence of crystals. These differences can be useful to distinguish the two species in culture.

The cultural studies are an important complement to the traditional taxonomy based upon basidiome morphology and, the enzimatic tests are an auxiliar tool to distinguish the type of rot. Nevertheless basidiomata can display individual behavior according to the conditions of the natural or the artificial environment. Thus the same species can give different responses, in nature or in culture, depending on the place, temperature and mainly the individual genetic characters.

Biochemical tests were negative in all the cases for tyrosinase (ρ-cresol) and guaiacol. Taylor (1974) performed biochemical tests with several species, in cultures of differents ages. In his work we could notice that the reactions to ρ-cresol and to guaiacol were observed only for cultures that were 10 days old or more while our cultures were seven days old when tests were performed.

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CONTRIBUTIONS À UN INVENTAIRE MYCOLOGIQUE DE MADAGASCAR. 2. NOUVEAUX TAXA DANS LE GENRE CANTHARELLUS

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ABSTRACT

Two new taxa within the genus *Cantharellus* were discovered during a recent mission in Madagascar. The very small *Cantharellus decolorans* spec. nov. is well-characterized by its pink colour becoming straw-yellow in the later stages. *C. platyphyllus ssp. bojeriensis* ssp. nov. differs from the type by its geographical localisation, its mycorrhizal association with the endemic woodland tree *Uapaca bojeri* and the paler colours.

RESUME

Une récente mission à Madagascar a permis de découvrir deux nouveaux taxons se rapportant au genre *Cantharellus* Fr.. Le tout petit *C. decolorans* spec. nov. est caractérisée par sa couleur rose devenant jaune paille dans l'âge. *C. platyphyllus ssp. bojeriensis* ssp. nov. se différencie du type par sa localisation géographique, son association mycorhizienne avec l'arbre endémique *Uapaca bojeri* et ses couleurs plus pâles.

INTRODUCTION

Patouillard (1924) a décrit les deux premières "chanterelles" malgaches: *C. madagascariensis* et *C. avellaneus*. Depuis, le genre *Cantharellus* n'a plus été étudié à Madagascar exception faite pour la publication par Heim (1936) d'une nouvelle espèce en provenance du littoral, *C. cyanoxanthus*. Invalidement introduit par absence de diagnose latine, ce taxon fut plus tard validé par Heinemann (1958).

Dans cette deuxième contribution à l'inventaire mycologique de Madagascar (la première ayant été publiée par Buyck, Eyssartier & Duhem, 1998), nous présentons deux nouveaux taxons de chanterelles des forêts indigènes dans lesquelles ils sont associés à diverses espèces du genre *Uapaca.*

MATÉRIEL ET MÉTHODES

Cette étude est basée sur du matériel récolté à Madagascar lors de deux missions du Muséum National d'Histoire Naturelle durant les mois de mars 1996 puis janvier et février 1997.

Les diamètres piléiques mesurés sur nos récoltes sont les diamètres projetés (Josserand, 1952).

Les couleurs sont codées en référence à Kornerup & Wanscher (1978). Les abréviations des herbiers sont d'après Holmgren et al. (1990), celles des auteurs d'après Brummit & Powell (1992)

Les observations microscopiques ont été réalisées sur des préparations colorées au rouge congo ammoniacal après un bref traitement à la potasse 5%. De petits fragments de pileipellis ont été systématiquement observés dans l'acide sulfurique (H_2SO_4) à 80%, afin d'observer un éventuel changement de couleur qui peut être en relation avec la présence de pigments caroténoïdes (Heim, 1949).

Les mesures sporales, établies pour 20 spores et données en micromètres (μm), comprennent les minima et maxima entre parenthèses et les moyennes en italique assorties de leur écarts-types. Le coefficient longueur/largeur (Q) est noté selon les mêmes règles.

Sur les planches de dessins, les barres d'échelle représentent 1 cm pour les caractères macroscopiques, 10 μm pour les caractères microscopiques.

DESCRIPTION DES ESPÈCES

Cantharellus decolorans Eyssartier & Buyck *sp. nov.*

Figs. 1-4.

Pileus (2) 6-15 µm diam., primo planus margine involuto dein valde infundibiliformis margine irregulare sinuoso lamellis modo striato ; pileipellis primo hirsuta mox sublaevis impolita pallide rosea flava straminea. Hymenophorum generaliter facile separatum, plicis irregularibus

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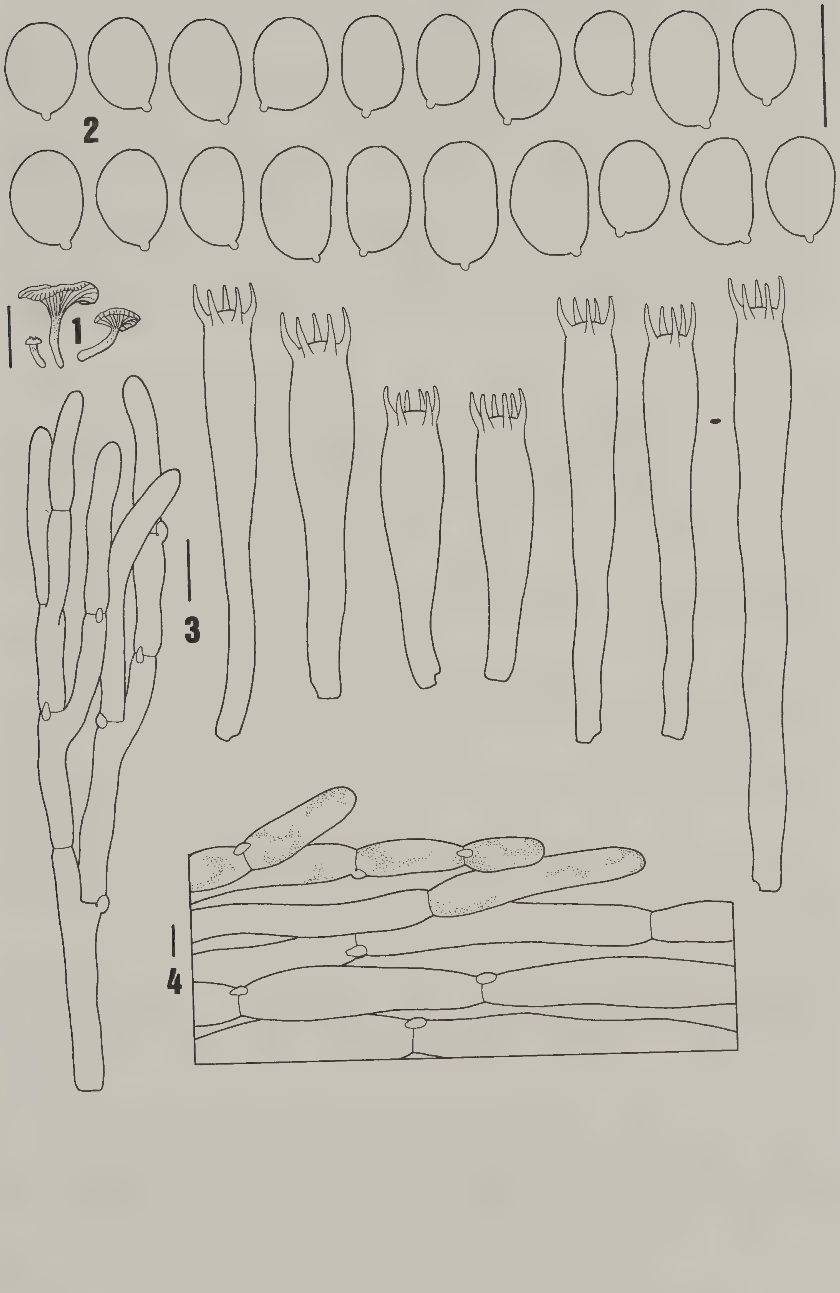
Figs. 1-4. *Cantharellus decolorans.* **1 .** Basidiomes **2 .** Spores **3 .** Basides et éléments hyméniens **4 .** Suprapellis.



distantibus frucato-anastomosantibus crassis longe decurrentibus pilei concoloris compositum. Stipes (5)10-15 x (0,5)1-1,5 mm, cilindratus, subtile fibrillosus, pilei concolorus, cavus in vetere. Caro tenuis, concolor, dulci, haud sensitiva actione AgNO3, H2SO4, TL4, gaiac.

Sporae (6,5) 6,79-7,58-8,37 (9) x (5) 5,2-5,63-6,06 (6,5) µm, Q = (1,08) 1,23-1,35-1,47 (1,67), breviter ellipsoideae vel subglobosae ut ovoideae, laeves, pallide flavae. Basidia dimensione variabilia, (42) 50-70 (95) x (7) 8-10 µm, cilindrato-clavata, 5-6 sporigera. Trama lamellarum paululum evoluta. Suprapellis hyphis (4) 8-13 (15) µm diam. cute formantibus extremitatibus liberis numerosis brevibus raro emergentibus efformata. Pigmentum intracellulare granuloso flavum actione acido sulfurico 80% brunnescens, saepe pigmento parietale laeve vel plus minusve incrustate accompagnato. Fibulae omnibus partis adsunt. Holotypus : Buyck-Eyssartier-Moreau 97227 (PC).

Pileus (2) 6-15 mm, d'abord plan à marge rabattue puis nettement infundibuliforme, à marge seulement un peu brisée, irrégulière, ondoyeuse et plus ou moins cannelée cabossée par l'empreinte des lames; revêtement hirsute sur les primordia puis très rapidement sublisse, mat, d'un beau rose pâle (7A4; 7A5; 8A4), devenant entièrement jaune à jaune paille sale dans l'âge (env. 4A5) - on trouve aussi de petits exemplaires apparemment mal venus et stoppés dans leur croissance qui possèdent déjà cette couleur -, les basidiomes passant par différents états où les deux couleurs coexistent. **Hyménophore** constitué de plis irréguliers, espacés, fourchus anastomosés et épais, longuement décurrents, concolores au pileipellis et se décolorant de la même façon, à arête obtuse concolore; l'hyménophore se détache généralement facilement de la chair du chapeau, au moins chez les plus gros exemplaires. **Stipe** (5) 10-15 x (0,5) 1-1,5 mm, égal, creux chez les plus gros exemplaires, très finement fibrilleux longitudinalement, concolore ou un peu plus foncé (7A6) et se décolorant comme les autres parties du basidiome. **Chair** très mince, concolore aux parties, à odeur et saveur très faibles, douces. **Réactions macrochimiques** nulles ou insignifiantes avec les réactifs suivants: KOH, sulfoformol, métol, $AgNO_3$, phénolaniline, phénol, formol, sulfate de fer, teinture de gaiac, TL4.

Exsiccatum montrant un revêtement piléique et un hyménophore concolores, beige jaunâtre à jaunâtre pâle, le stipe étant clairement discolore et plus foncé, brun rougeâtre, orangé rougeâtre ou beige rosâtre.

Spores (6,5) 6,79-*7,58*-8,37 (9) x (5) 5,2-*5,63*-6,06 (6,5) µm, Q = (1,08) 1,23-*1,35*-1,47 (1,67), courtement ellipsoïdes à subglobuleuses ou un peu ovoïdes de face, lisses, à cytoplasme assez dense, finement émulsionné ou parfois guttulé, jaunâtre pâle. **Basides** très variables de taille y compris dans un même basidiome, (42) 50-70 (95) x (7) 8-10 µm, cylindro-clavées, optiquement vides ou finement guttulées, 5-6 sporiques. **Trame lamellaire** très réduite. **Suprapellis** un cutis d'hyphes de (4) 8-13 (15) μm diam., à extrémités libres nombreuses rarement un peu redressées, souvent courtes et plus ou moins articulées. **Pigment** majoritairement intracellulaire sous la forme d'un précipité jaunâtre qui brunit dans l'acide sulfurique à 80%, souvent accompagné d'un pigment pariétal lisse à plus ou moins incrustant par endroit. **Boucles** présentes dans toutes les parties du basidiome.

Matériel examiné : Province de Tamatave : station forestière de La Mandraka, forêt dense humide d'altitude, sous *Uapaca densifolia*, en troupe dans une souche creuse, 3-II-1997, Buyck-Eyssartier-Moreau 97227 (holotypus PC). *Ibid.*, 14-II-1997, Buyck-Eyssartier-Moreau 97494 (PC). *Ibid.*, 20-II-1997, Buyck-Eyssartier-Moreau 97585 (PC), réserve biologique d'Andasibe, forêt dense humide d'altitude, en troupe dans un tronc creux de *Pandanus*, 13-II-97, Buyck-Eyssartier-Moreau 97463 (PC).

Distribution : endémique.

Observations : parmi les espèces affines, *C. roseus* (Schw.) Fr., espèce mal connue décrite de Caroline du Nord, en diffère au moins par son hyménophore "rugueux, blanc". *C. carneoflavus* décrite par Corner (1969) de Bornéo diffère par sa taille plus importante, sa chair immuable et ses spores nettement plus grandes.

C. omphalinoides Corner, *C. diminutivus* Corner et *C. schmitzii* Heinem., dont les basidiomes présentent une taille et une microscopie équivalentes, ne possèdent aucun pigment rose.

C. decolorans semble proche de *C. ruber*, décrite par Heinemann (1966) du Zaïre, à la fois par sa morphologie générale, sa décoloration, ses boucles abondantes et son pigment intracellulaire brunissant dans l'acide sulfurique. *C. ruber* est cependant bien caractérisée par ses basidiomes beaucoup plus grands et de couleur différente, microscopiquement par ses spores nettement plus larges.

Il est intéressant de constater que nous avons toujours récolté cette espèce dans de vieilles souches partiellement détruites. La croissance et la fructification de champignons appartenant à des groupes réputés ectomycorhiziens sur le bois ou sur des débris végétaux ne sont pas exceptionnelles en régions tropicales humides. Elles ne sont même pas rares en régions arctiques (Buyck, obs. pers.). Indiquant probablement des capacités saprotrophiques accrues, ce phénomène mériterait de faire l'objet d'études approfondies.

Cantharellus platyphyllus **Heinem.** ***ssp. bojeriensis*** Eyssartier & Buyck *ssp. nov.*

Figs. 5-8.

A typo praesertim differt distributione Madagascariense Uapacae bojerii in silva clariore consociato coloribus pallidioribus (pileo aurantiaco aurantiaco-flavo rubro-aurantiaco locale lazulino griseo-lazulino vel glauco-veneto tincto stipite pallidiore eisdem coloribus tincto). Holotypus : Arivonimamo, ad terram in silva clariore sub Uapacae bojerii, 30-I-1997, Buyck-Eyssartier-Moreau 97055 (PC).

Pileus (20) 25-45 mm, plan convexe à plan, à marge rabattue ou plus ou moins lobée-difforme; revêtement lisse, le plus souvent ridé-rugueux surtout au centre, de couleur orange vif à orange jaune vers la marge (4A8; 4A7), souvent à reflets rougeâtres au disque (lavé de 9A7), avec des reflets bleus, gris bleuté ou glauques et caractéristiques (lavé de 20B2, 20C2), rarement totalement absents. **Hyménophore** formé de lames espacées, larges et bien formées, 3-5 mm de large, de différentes longueurs, crispées-anastomosées au fond des sini, largement décurrentes, peu fourchues; couleur crème jaunâtre assez pâle (env. 4A3), à peine plus orange vers la marge, souvent avec un reflet glauque, à arête entière concolore. **Stipe** 25-40 x 8-10 mm, subégal, plein, jaune orangé pâle subconcolore aux lames, lisse. **Chair** assez épaisse, colorée de jaune sous les revêtements, très nettement blanc bleuté chez les jeunes, rarement blanche seulement un peu glauque; odeur et saveur agréables, comme *C. cibarius* en plus fruités. **Réactions macrochimiques** nulles ou insignifiantes avec les réactifs suivants: KOH, sulfoformol, métol, $AgNO_3$, phénolaniline, phénol, formol, sulfate de fer, teinture de gaiac, TL4.

Exsiccatum entièrement brun jaunâtre terne, le revêtement piléique ne gardant que rarement quelques traces de rose ou de brun rougeâtre.

Spores (7,5) 7,91-*8,73*- 9,55 (10,5) x (5,5) 5,94-*6,43*-6,92 (7) μm, Q = (1,23) 1,28-*1,36*-1,44 (1,5), largement ellipsoïdes à parfois subglobuleuses dans les petites longueurs, lisses, à cytoplasme dense, un peu jaunâtre, en général homogène, parfois guttulé. **Basides** (2) 4-5 sporiques, (50) 55-65 (76) x 8-10 μm, cylindro-clavées, optiquement vides ou guttulées. **Trame** emmêlée formée d'hyphes grêles 3-5 μm et d'hyphes plus larges jusqu'à 10 (12) μm de diamètre. **Suprapellis** en cutis d'hyphes mesurant (3) 5-10 (15) μm de diamètre, plus ou moins articulées. **Pigment** nettement intracellulaire, sous forme de gouttelettes huileuses, souvent plus ou moins

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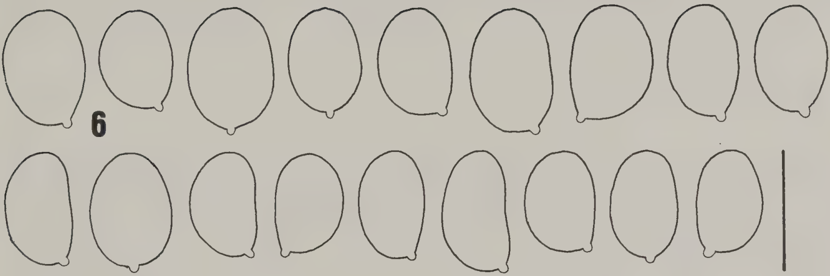
Figs. 5-8. *Cantharellus platyphyllus* ssp. *bojeriensis.* **5 .** Basidiomes **6 .** Spores **7 .** Basides **8 .** Suprapellis.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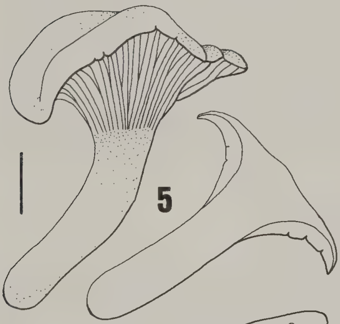
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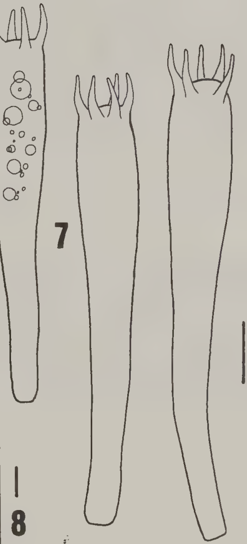
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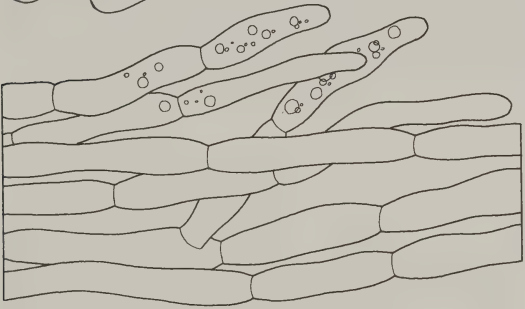
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précipitées, jaune-orange, qui brunissent dans l'acide sulfurique à 80%. **Boucles** absentes dans toutes les parties du basidiome.

Matériel examiné : Province d'Antananarivo : Arivonimamo, forêt claire sous *Uapaca bojeri,* 30-I-1997, Buyck-Eyssartier-Moreau 97055 (holotypus, PC). *Ibid.*, 30-I-1997, Buyck-Eyssartier-Moreau 97079 (PC, TAN), 97087 (PC). *Ibid.*, acheté sur le marché à Antananarivo, 20-III-1996, Buyck 6472 (PC), 6473bis (PC, TAN).

Distribution : endémique.

Observations : cette sous-espèce se différencie du type, décrit du Zaïre, par ses couleurs différentes, sa distribution géographique et son association avec un arbre endémique de Madagascar, *Uapaca bojeri.* Elle fait partie d'un groupe d'espèces proches africaines, comprenant *C. symoensii* Heinem. et *C. splendens* Buyck, bien caractérisées par leurs couleurs vives et leurs hyphes toujours afibulées. Si *C. symoensii* et *C. splendens* montrent, au niveau des hyphes du suprapellis, des paillettes intracellulaires rouge vif qui bleuissent fortement au contact de l'acide sulfurique, nous n'avons jamais pu mettre en évidence cette réaction sur aucune de nos récoltes de *C. platyphyllus sensu lato*, qu'elles soient africaines ou malgaches.

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THE NOMENCLATURAL PROPRIETY OF RHIZOPHLYCTIS ROSEA

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ABSTRACT

Nomenclature of the chytridiomycete *Rhizophlyctis rosea* as part of the *Rhizophlyctis-Karlingia-Karlingiomyces* morphological assemblage is reexamined. Nomenclatural problems in this group of genera center around the original description of *Karlingia rosea*, a common soil chytrid. The genus *Karlingia* was based in the original description exclusively on *Rhizophlyctis rosea* and initially contained only *K. rosea*. *Karlingia* and *K. rosea* are nomenclaturally superfluous and should be rejected in favor of *Rhizophlyctis* and *R. rosea,* the taxon already designated as the type of *Rhizophlyctis*. Only *Rhizophlyctis* and *Karlingiomyces* are available as legitimate generic names for species belonging to this morphological assemblage of chytrids.

NOMENCLATURAL CHRONOLOGY AND ANALYSIS

Fischer (1892) established the genus *Rhizophlyctis* to include chytrids with eucarpic, monocentric thalli and with rhizoids originating from several points on extramatrical sporangia. The thallus is characteristically interbiotic and only the distal portions of the rhizoidal axes are embedded in the substrate. Fisher (1892) recognized five species of *Rhizophlyctis* in the following order: *R. braunii* (Zopf) Fischer, *R. vorax* (Strasburger) Fischer, *R. mastigotrichis* (Nowakowski) Fischer, *R. mycophila* (Braun) Fischer, and *R. rosea* (de Bary & Woronin) Fischer. These species were transferred to *Rhizophlyctis* from either *Rhizidium* or *Chytridium*, and were accorded equal status by Fischer. An illustration (based on a drawing by Nowakowski) was provided for *R. mastigotrichis,* but this species was not otherwise specially preferenced by Fischer. *Rhizophlyctis mycophila* has the oldest epithet, as *Rhizidium mycophilum* Braun (1856); it is the type of the genus *Rhizidium* Braun (cf. Sparrow, 1960), and is unavailable under *Rhizophlyctis*. *Rhizophlyctis braunii* and *R. vorax* also belong to the genus *Rhizidium* (Sparrow, 1960). *Rhizophlyctis rosea* (based on *Chytridium roseum* de Bary & Woronin,1865) is the species still available under *Rhizophlyctis* with the oldest epithet.

The species of *Rhizophlyctis* included by Fischer (1892) are potentially syntypic. Lacking a holotype or isotype, these syntypes would all be available for lectotype selection (Articles 9.9, 10.1, 10.2, ICBN, 1994), except, as indicated, *R. braunii*, *R. vorax* and *R. mycophila* are morphologically placed in *Rhizidium* not *Rhizophlyctis* (Sparrow, 1960). Hence, *Rhizophlyctis rosea* and *R. mastigotrichis* are the syntypes from which a supportable lectotype for *Rhizophlyctis* could have been chosen. Syntypic situations are common prior to instigation of the type method in nomenclature in 1930 (cf. Weatherby, 1949). Clements and Shear (1931) listed the type of *Rhizophlyctis* as *R. rosea*, in effect, designating *R. rosea* as lectotype of the genus. It is in the domain of semantics that, technically, the type of *Rhizophlyctis* is whatever material (most certainly perished) might have been conjurable as the type of *R. rosea* (Article 10.1, ICBN, 1994). By the wording of Article 9.2, de Bary and Woronin's (1865) illustration of *Chytridium roseum* (Plate 2, Figs. 17-20) may be accepted as the type of *R. rosea.*

Johanson (1944), based primarily on endo-operculation versus exo-operculation or inoperculation of exit papillae of sporangia of rhizophlyctoid taxa, described the new genus *Karlingia*. Johanson observed sporangial papillae of the *Rhizophlyctis rosea* material she examined to be consistently endo-operculate, in contrast to previous descriptive information (citing, for example, Ward, 1939). Johanson also discussed minor putative differences in zoospore pigmentation, cytoplasmic density, and "granule" number. Bernstein (1968), who did not cite Johanson's work, documented considerable variation in zoospores of *Rhizophlyctis rosea*, particularly in size and number of lipoid globules. Regardless of the consistency of endo-operculation of papillae (or other morphological features), nomenclaturally, Johanson based *Karlingia* (and *K. rosea*) solely on *Rhizophlyctis rosea* (*Chytridium roseum*); no other species were included in *Karlingia* when it was described. It would have been nomenclaturally preferable had Johanson merely emended the description of *Rhizophlyctis rosea* to reflect her observations on morphology.

Sparrow (1960) considered endo-operculation of exit papillae an unstable character, of sporadic occurrence in various chytrids (e.g., *Diplophlyctis*, *Nephrochytrium*). He stated that endo-operculation may even occur in "the true operculate [exo-operculate] as well as the inoperculate types." Sparrow (1960), based on his observations and those of Haskins (1948), concluded that some "strains" of *Rhizophlyctis rosea* exhibit endo-operculation, whereas others do not. Sparrow noted that the original figures and description of *Chytridium roseum* (de Bary and Woronin, 1865) provide evidence of endo-operculation. Indeed, de Bary and Woronin's Figure 18a (Plate 2) illustrates what is apparently such a structure. On morphological grounds, Sparrow (1960) regarded *Karlingia* Johanson as a synonym of *Rhizophlyctis* Fischer, and described the new genus *Karlingiomyces* to accommodate definitively exo-operculate species of the rhizophlyctoid assemblage *(Rhizophlyctis-Karlingia-Karlingiomyces)*.

Dogma (1973) concluded that characters of endo-operculation, exo-operculation, and inoperculation are generically meaningful in the rhizophlyctoid assemblage. To summarize, Dogma suggested the following system: *Karlingiomyces* would accommodate exo-operculate species; *Karlingia* would encompass endo-operculate taxa; and *Rhizophlyctis* would be reserved for inoperculate organisms. Dogma considered the morphology of endo-operculation to be of sufficient complexity to perhaps warrant subgeneric distinctions within *Karlingia*. Of nomenclatural concern is that Dogma designated *Rhizophlyctis mastigotrichis* as the lectotype of the genus *Rhizophlyctis*, with no mention of the prior typification by Clements and Shear (1931).

In the rhizophlyctoid composite of genera, Karling (1977) recognized only *Rhizophlyctis* Fischer (1892) and *Karlingia* Johanson (1944). Karling (1977) treated *Karlingiomyces* Sparrow (1960) as a synonym of *Karlingia.* Karling's rationale was that of placement of all operculate species of this morphological assemblage (albeit exo- or endo-operculation) in one genus, *Karlingia* (which would, by this logic, have priority over *Karlingiomyces*). Karling, citing also observations of other workers such as Chambers and Willoughby (1964), viewed endo-operculation of exit papillae of *Karlingia rosea* (which he considered possibly "most common of all chytrids") as a more stable feature than did Sparrow (1960). Karling (1977), as did Dogma (1973), used the name *Rhizophlyctis* for inoperculate taxa of the complex. Dogma (1973) and Karling (1977) provided valuable discussions on generic concept development for *Rhizophlyctis* (e.g., its distinction from *Rhizidium* based on number of main rhizoidal axes).

Longcore (1996) correctly noted that whereas Dogma (1973) selected *R. mastigotrichis* as lectotype of *Rhizophlyctis*, typification of the genus had already been accomplished by Clements and Shear (1931) when they listed *R. rosea* as the type of *Rhizophlyctis*. Longcore (1996) enumerated all three genera of the rhizophlyctoid assemblage (*Rhizophlyctis*, *Karlingia*, and *Karlingiomyces*), stating that *Karlingiomyces* Sparrow was rejected by Karling (1966), but accepted by Dogma (1973). Longcore, however, did not offer an opinion on generic synonymy within this assemblage.

Based on previous findings (e.g., Powell, 1976), we agree with Sparrow (1960) that endo-operculation is a variable feature, perhaps inconsistent for general use in the generic taxonomy of chytrids. Electron microscopy confirms Sparrow's (1960) report that the endo-operculum is not necessarily dehisced (Powell, 1994; fig. 52). The recognition of *Karlingia* Johanson on morphological grounds is thus debatable, although there is no dispute that the question of operculation is significant in the rhizophlyctoid assemblage. But, whether or not one accepts *Karlingia* morpho-taxonomically, nomenclatural regulations override systematic considerations in determining synonymy. Two nomenclatural points bear directly on this determination. The first point of nomenclature is that Clements and Shear (1931) *de facto* lectotypified *Rhizophlyctis* with *R. rosea*. This occurred after formal installment of the type method (1930), and 42 years before Dogma's stated lectotypification of *Rhizophlyctis* with *R. mastigotrichis*. If a substantial basis for overturning a lectotypification cannot be shown, then the original authors' choice is to be followed (Article 9.13, ICBN, 1994). We find no reason of substance to overturn the type selection by Clements and Shear (1931). Thus, *Rhizophlyctis rosea* (based on *Chytridium roseum*) is the lectotype (and determines application) of the name *Rhizophlyctis*. The second nomenclatural point concerns Johanson's (1944) publication of *Karlingia*. Although considering her isolates somewhat different from others of the same taxon, Johanson nonetheless established *Karlingia* (and its only species, *K. rosea*) solely on the previously and validly described genus and species, *Rhizophlyctis* and *R. rosea*. This constitutes creation of a nomenclaturally superfluous genus (and species) name (McVaugh et al., 1968; Article 52.1, 52.2, ICBN, 1994). The name *Karlingia* Johanson should thus be rejected on nomenclatural grounds, and treated as an illegitimate synonym of *Rhizophlyctis* Fischer. *Karlingia* should not be considered available as a legitimate generic name for any chytrid.

In summary the nomenclatural situation for the *Rhizophlyctis-Karlingia-Karlingiomyces* assemblage is as follows. *Karlingia* may not legitimately be used as a generic name for a chytrid, or for any fungus or plant (cf. ICBN, 1994: Preamble statement 7; Articles 6.4 and 52). *Karlingiomyces* Sparrow is available nomenclaturally for consistently exo-operculate species of the morphological assemblage. Remaining taxa of the assemblage would seemingly be referred (by morphology or by default) to the genus *Rhizophlyctis* Fischer, including inoperculate and any presumably endo-operculate species. Thus, the species of *Karlingia* additional to *K. rosea* listed by Dogma (1973) and Longcore (1996) are tentatively remanded to *Rhizophlyctis*. However, we are not formally transferring these *Karlingia* species to *Rhizophlyctis* because, to be certain, the organisms should be reisolated and studied, especially the details of their zoospore architecture (cf. Barr, 1980). Zoospores examined ultrastructurally in the *Rhizophlyctis-Karlingia* complex are of the spizellomycetalean type or are of uncertain affinity (Barr, 1980; Barr and Desaulniers, 1986). However, the zoospore of *Rhizophlyctis harderi* Uebelmesser (1956) is chytridialean in character (Powell and Roychoudhury, 1992). Thus, in the rhizophlyctoid assemblage, the question becomes, to which order a species belongs not simply to which genus.

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BONDARCEVOMYCES, A NEW GENUS OF POLYPORES (HYMENOMYCETES, BASIDIOMYCOTA)

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Abstract

A new genus, *Bondarcevomyces* Parmasto is described and a new combination, *B. taxi* (Bondartsev) Parmasto is proposed. The relationships with other genera of Aphyllophorales are discussed.

Key words: Bondarcevomyces, polypores, Phaeolaceae, Aphyllophorales, taxonomy.

Bondarcevomyces Parmasto, **gen. nov.**

Basidioma pileatum, molle, caseosum et aquosum (in sicco fragile et levissimum), aurantiacum, rubro-aurantiacum vel subfuscum; tubuli tactu olivascens. Odor fortis, suavis, basidioma etiam in sicco valde foetens. Rhizomorphae desunt. Systema hypharum monomiticum, hyphae generatoriae tenuiter tunicatae, fibulatae. Hyphae gloeoplerae adsunt, cystidia desunt. Basidia clavata, 4 sterigmatis. Basidioporae crassiuscule tunicatae, flavae vel brunneae, dextrinioideae. Massa sporarum ochraceum vel brunneum. Putredo ligni destructiva (brunnea).

Typus: *Hapalopilus (Polyporus) taxi* Bondartsev, Bot. Mater. Otd. Sporov. Rast. Bot. Inst. Akad. Nauk S.S.S.R. 5 (1-3): 17, 1940 ≡ *Bondarcevomyces taxi* (Bondartsev) Parmasto.

Etymology. *Apollinari S. Bondarzew* and his daughter *Margarita A. Bondarceva*, Russian mycologists, authors of numerous works on Polyporaceae s.l.; *μύκης*, fungus. Gender: m.

Basidiomata pileate, soft, brittle and very light when dry, pilear surface orange, reddish orange or brown; tubes turning olive when bruised; smell strong, sweet, persistent in herbarium; rhizomorphs absent. Hyphal system monomitic; generative hyphae thin-walled, with clamps; gloeoplerous hyphae present, cystidia absent; basidia clavate, with 4 sterigmata; basidiospores with smooth, thickened, yellow or brownish dextrinoid walls; spore print ochraceous or brown(ish). Causing brown rot of wood.

Type of the new genus *H. taxi* has been combined in *Tyromyces* and *Parmastomyces*. All these genera have hyaline spores. *Hapalopilus* and *Tyromyces* species have indextrinoid and thin-walled spores; they cause a white rot of wood; gloeoplerous hyphae are absent in the basidiomata of *Hapalopilus* and most species of *Tyromyces*. The pigment causing brownish colour in *B. taxi* is different from the pigment(s) of *Hapalopilus* spp. (cf. Figs. 1-2). *Parmastomyces* has basidiomata with different colour and colour changes, a dark gelatinous line above the tube layer, lacks gloeoplerous hyphae, and has hyaline spores.

The peculiar colour and its change, as well as the consistency in *B. taxi* are also typical of *Phaeolus schweinitzii* (Fr.: Fr.) Pat., which has gloeocystidia and also causes brown rot of wood. The colour of *Phaeolus* is caused by different pigments (Fig. 3), and its spores are hyaline and indextrinoid. *Hapalopilus olivascens* Corner has similar colour and

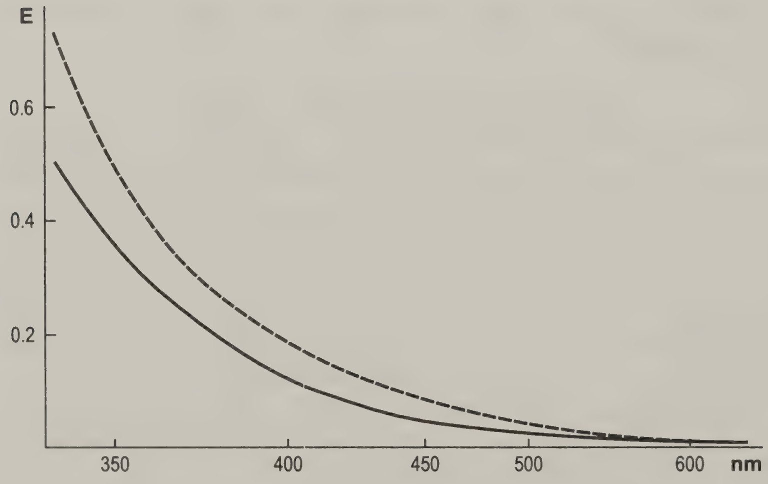

Fig. 1. *Hapalopilus croceus.* Absorption spectrum of aqueous pigment extract and alkalized aqueous extract (broken line). Methods used: see Parmasto & Parmasto, 1979: 206.

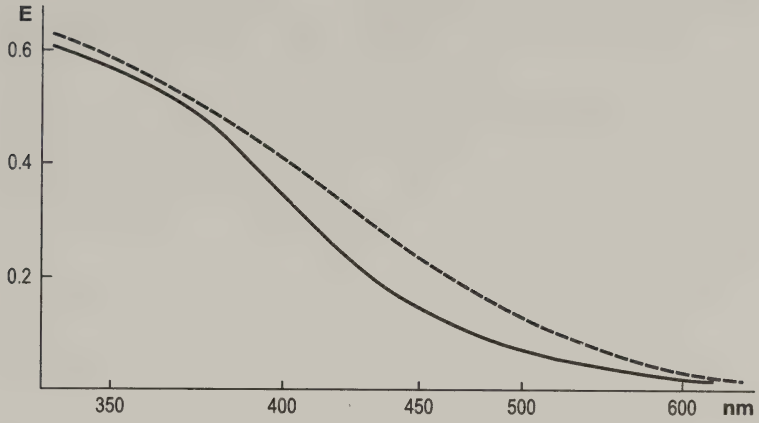

Fig. 2. *Bondarcevomyces taxi.* Absorption spectrum of aqueous extract and alkalized aqueous extract (broken line).

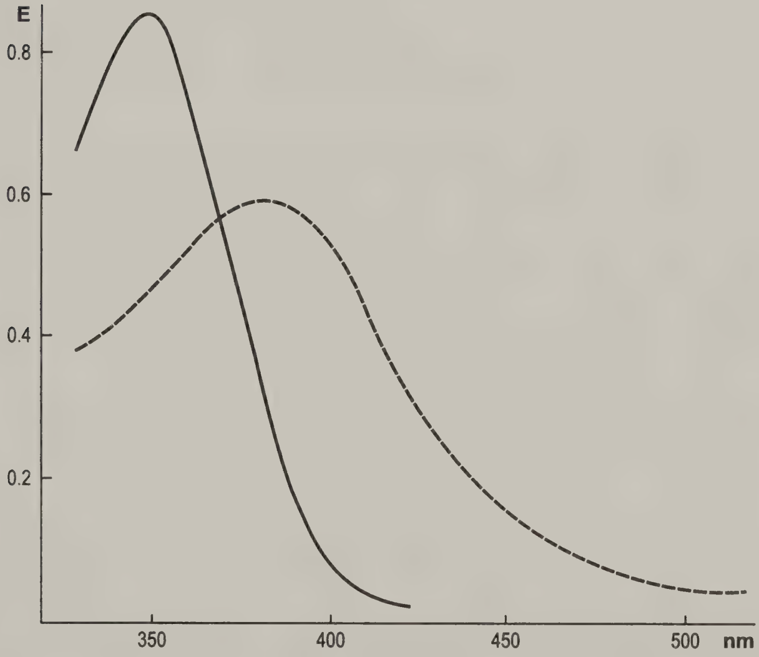

Fig. 3. *Phaeolus schweinitzii.* Absorption spectrum of aqueous extract and alkalized aqueous extract (broken line). (From: Parmasto & Parmasto, 1979, fig. 8.)

colour change of basidiomata (see Corner, 1989: 69), but differs, *inter alia*, by its strongly inflated hyphae and white, thin-walled indextrinoid spores.

There are only few genera of polypores with brownish spores. *Loweporus* Wright has a dimitic hyphal system without gloeoplerous hyphae; *Phaeotrametes* Wright has a trimitic hyphal system, inamyloid, indextrinoid spores, and gloeoplerous hyphae are absent in its only species (Wright, 1966). The only genus possibly related to *Bondarcevomyces* is *Pseudopiptoporus* Ryvarden with its only species *P. devians* (Bres.) Ryvarden. The latter species has been found only once in Mozambique on angiospermous wood (cf. Ryvarden & Johansen, 1980). It differs mainly from *Bondarcevomyces* in its dimitic hyphal system with generative hyphae and amyloid binding hyphae.

The differences indicated above show that *Bondarcevomyces* has an unique combination of important characters in polyporoid fungi. Also, there are no corticioid fungi with this unique combination. All this leaves *Bondarcevomyces* without a distinct place in the classification of Aphyllophorales. Temporarily it may be united with *Phaeolus* into Phaeolaceae (cf. Fiasson & Niemelä, 1984: 22), but not including that family into Hymenochaetales as has been done by Fiasson & Niemelä.

Bondarcevomyces taxi (Bondartsev) Parmasto, **comb. nov.**

Basionym: *Hapalopilus (Polyporus) taxi* Bondartsev, Bot. Mater. Otd. Sporov. Rast. Bot. Inst. Akad. Nauk S.S.S.R. 5 (1-3): 17, 1940.

Synonyms: *Tyromyces taxi* (Bondartsev) Ryvarden & Gilb., European Polypores 2: 693, 1994; *Parmastomyces taxi* (Bondartsev) Y.-C. Dai & Niemelä, Ann. Bot. Fennici 32: 222, 1995.

Figures and an additional description: Ryvarden & Gilbertson, 1994, f. 379; Dai & Niemelä, 1995, f. 5; Chang, 1996: 34, f. 12.

Rem.: Colour terminology used below is as in Kornerup & Wanscher's (1967) handbook; colour notations are as in the Munsell Book of Color (1942).

Basidiomata 3-8 x 2-12 x (0.7)-1-2.5 cm, annual, sessile, dimidiate, flabellate, thickening towards base, usually a few imbricated, rarely single, cheesy, fleshy or spongy, watery, drying brittle and very light. Odour strong, cloying sweetish, in dry specimens very strong, almost unpleasantly cloying honey-like. Pilear surface slightly floccose, soft, sometimes indistinctly zonate, when fresh orange red, brownish orange, yellowish brown, later almost tomato red or dark rust brown (Munsell: 2.5 YR - 7.5 YR 4/10, sometimes 7.5 R 3/8), near margin dark yellow to orange red (5 YR 6/13 or 7.5 R 5/14), later margin concolorous, thin, rounded or almost sharp. Pilear surface of herbarium specimens brick red or reddish brown (2.5 - 5 YR 5-6/8-10 or 4-5/6-8). Context thick, indistinctly zonate or azonate, ochraceous, light orange to almost carrot red (7.5 - 2.5 YR 6-7/12-14, later sometimes 2.5 YR 4/10), turning dark greenish or olivaceous when bruised (5 Y 5/4); in herbarium specimens butter yellow or maize yellow (2.5 Y 8/8-10, sometimes 2.5 Y 9/6), blackish in KOH solution. Tubes 3-7 mm long, cheesy or fleshy, light orange or melon yellow (10 YR 7/12 to 8/8), when bruised turning slowly olive green or honey yellow (2.5 Y 5/6); in herbarium specimens brittle, olive yellow or olive (2.5 Y 6-7/8-10) or blackish. Pores angular or slightly irregular, (0.15)-0.2-0.4-(0.6) mm in diam., (2)-3-(4) per mm, with rather thick dissepiments, pore surface light or deep orange to saffron yellow (10 YR - 2.5 Y 8-8.5/11-14), when bruised turning slowly olive (2.5-5 Y 5/6 to 5 4/4), in herbarium specimens dark olive to blackish (2.5 Y 5-6 to 5 Y 4/2-4). Spore print yellow ochre or ochraceous brown (10 YR 5-6/8-10), not changing in herbarium during many years.

Hyphal system monomitic. Generative hyphae sparingly branched, with anastomoses, thin-walled, with clamps, hyaline, (2.5)-3-5.5-(8) µm in diam., in context intermingled, in tubes subparallel and encrusted with olivaceous yellowish granules soluble in KOH solution. Gloeoplerous hyphae present, usually more abundant in tubes, slightly tortuous, sparingly branched, thin-walled, with a milky greyish or olivaceous yellow content, with rare clamps, 3-10-(15) µm in diam. Subhymenium thin, not thickening. Cystidioles or hyphidia few, (3)-4-8 µm in diam. Basidia clavate, with a basal clamp, 15-20-(22) x 5.5-7 µm, with 4 sterigmata 4-5.5 µm long. Basidiospores ellipsoid, with one side flattened, or short cylindric, with thickened smooth yellowish walls, dextrinoid, mostly with one or several guttula, (3.5)-3.8-4.8-(5.2) x 2.4-2.8-(3) µm.

Causing brown cubical heart rot of living tree trunks and stumps.

Substrata. On trunks of living and dead trees of coniferous families Taxaceae, Pinaceae and Cupressaceae *(Chamaecyparis formosensis, Larix dahurica, Larix* sp., *Pinus koraiensis, Taxus baccata)*.

Distribution. Asia: Russia, at East coast of Black Sea in Caucasus; Russian Far East, Primorsk Terr.; Northeastern China (Chilin Prov.); Taiwan, Ilan Co.

Specimens studied: RUSSIA, Krasnodarsk Terr., Khosta near Sochi, on a stump of *Taxus baccata*, 20 Sep 1936 L. Vasilyeva (LE, holotype); Russian Far East, Primorsk Terr., Sikhote-Alin Biosphere Reserve, Ust'-Serebryanka, on base of a trunk of *Larix dahurica* in an *Abies* forest, 14

Sep 1987, U. Kõljalg (TAA 150026); nearby on trunks of *Pinus koraiensis*, 16 Sep 1987, U. Kõljalg (TAA 150118, 150120); Sikhote-Alin Biosphere Reserve, Maisa, on base of a *Larix dahurica* trunk in an Abies forest, 11 Sep 1990, I. Parmasto (TAA 150941); nearby, on a trunk of *Larix dahurica*, 13 Sep 1990, E. P. (TAA 151005); nearby, on base of a *L. dahurica* trunk, 16 Sep 1990, E. P. (TAA 151114); Siberia, South East part of Novosibirsk Reg., Salair, on a *Larix* stump, E. Zhukov (TAA 166178). CHINA, Changbai Forest Reserve, on base of a *Larix* tree, 10 Sep 1993, Y.-C. Dai 2058 (H; TAA 166337).

Remarks. According to Chang (1996), *B. taxi* causes white rot, according to Ryvarden & Gilbertson, 1994: 694, the rot is "presumably white". Specimens seen by me in nature caused brown heart rot of standing trees or stumps. Several herbarium specimens have been collected without substrate, but in two cases (TAA 150026 and 151005) typically brown-rotten wood is seen at the base of basidiomata.

The spores of *B. taxi* have been described as hyaline by all authors except Chang (1996: 34), who called them "hyaline to slight yellowish". We have obtained three spore prints in nature; these are all ochraceous brownish and did not change remarkably during the last eleven years. The reddish brown dark colour of older basidiomata is caused by abundant spores deposited on these during sporulation. This phenomenon is well known in polypores *Fomes fomentarius* and *Ganoderma applanatum.* The striking white or chocolate brown colour resp. of their basidiomata in sporulation periods is caused by thick cover of spores, fallen on their upper surface due to certain aerodynamical processes.

The spores of *B. taxi* are less variable than in most other polypores. Mean measurements in μm and mean length/width ratio *Q* (n = 30) are:

TAA 166178 (Centr. Siberia)	3.99	2.57	1.56
H, Dai 910 (China)	4.31	2.58	1.67
TAA 151005 (Russ. Far East)	4.41	2.58	1.71 (basidiome)
	4.65	2.67	1.74 (spore print)
TAA 150120 (Russ. Far East)	4.45	2.59	1.72
LE, holotype (Caucasus)	4.45	2.65	1.68
TAA 150941 (Russ. Far East)	4.47	2.59	1.73
TAA 150118 (Russ. Far East)	4.53	2.61	1.74
TAA 151114 (Russ. Far East)	4.57	2.65	1.73
TAA 150026 (Russ. Far East)	4.62	2.79	1.66

According to these data, *species* mean is 4.45 x 2.63 µm, *Q* = 1.69. Further, 90% expected *specimen* means (see Parmasto & Parmasto, 1987) are in limits 4.09-4.81 x 2.50-2.76 µm; *Q* = 1.58-1.80.

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ADDITIONS TO THE LICHEN FLORA OF THAILAND

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Abstract: Fifteen lichen species are reported for the first time from Thailand. *Laurera meristosporoides* P. M. McCarthy & Vongshewarat sp. nov. (Trypetheliaceae) and *Porina erawanensis* P. M. McCarthy & Vongshewarat sp. nov. (Trichotheliaceae) are described.

INTRODUCTION

Although the lichen flora of Thailand is highly diverse, until quite recently it was largely neglected, with the main published contributions sometimes appearing decades apart (Vainio 1909, 1921; Paulson 1930; Sato 1962; Yoshimura 1978). However, the 1990s saw a greatly increased interest in and appreciation of the lichen flora, especially among Thai botanists. Intensive, localised investigations were undertaken (Mongkolsuk *et al.* 1996; Boonpragob *et al.* 1998), and detailed studies of the responses of forest lichens to environmental change yielded valuable floristic and ecological information (Wolseley 1997; Wolseley & Aguirre-Hudson 1991, 1997a, b; Wolseley *et al.* 1994).

In this paper we document two new species of *Laurera* Rchb. (Trypetheliaceae) and *Porina* Ach. (Trichotheliaceae) and new records of 13 other lichens (in *Conotrema*, *Laurera*, *Polymeridium*, *Porina*, *Pyrenula* and *Trypethelium*).

THE SPECIES

1. Conotrema lumbricoides Sipman, *in* A. Aptroot *et al.*, *Biblioth. Lichenol.* 64: 52 (1997)

This lichen was recently reported from lowland forest in Papua New Guinea (Aptroot *et al.* 1997). The Thai specimen was collected in the far south of the country and agrees closely with the well-illustrated protologue.

SPECIMEN EXAMINED: *Nakhon Sri Thammarat*: Khao Luang National Park, 8°22'N, 99°40'E, alt. 300 m, on bark in evergreen forest, 17.ii.1998, *K. Vongshewarat* (RAMK 9932).

2. Laurera keralensis D. K. Upreti & A. Singh, *Bull. Jard. Bot. Belg.* 57: 374 (1987)
Thallus yellowish brown, smooth; photobiont *Trentepohlia*. Ascomata dark brown to black, 0.3–1 mm diam., solitary or aggregated in irregular pseudostromata. Ascomatal

apex convex, cracked, 0.1–0.25 mm diam. Hymenium comprising anastomosing pseudoparaphyses and elongate, 8-spored asci bathed in a clear oily substance. Ascospores elongate-ellipsoidal, muriform, with 9–12 transverse divisions, 50–60 × 15–20 μm.

This lichen was previously known from Kerala, India (Upreti & Singh 1987).

SPECIMENS EXAMINED: *Nakhon Ratchasima*: Khao Yai National Park, 14°11'N, 101°11'E, alt. 1400 m, on bark in evergreen forest, viii.1997, *K. Boonpragob et al.* (RAMK 8458). *Sakonnakhon*: Ratchapat Sakon, 16°53'N, 104°26'E, on bark of *Mangifera indica*, 19.v.1998, *W. Khumtim 371* (RAMKH).

3. Laurera megasperma (Mont.) Riddle, *Bull. Torrey Bot. Club.* 44: 323 (1917)
Thallus pale yellowish green, smooth; photobiont *Trentepohlia*. Ascomata solitary or paired in 0.8–1.5 mm diam. pseudostromata that are prominent and largely concolorous with the thallus, or immersed in the substratum. Ascomatal apex plane to slightly convex, blackish, 0.4–0.75 mm diam., with an external, whitish ring. Hymenium comprising anastomosing pseudoparaphyses and elongate 4-spored asci bathed in a clear, oily substance. Ascospores elongate-cylindrical, muriform, with 40–65 transverse divisions, 180–320 × 28–34 μm.

This lichen is known from the Neotropics, India and Sri Lanka (Letrouit-Galinou 1957, Makhija & Patwardhan 1988).

SPECIMENS EXAMINED: *Nakhon Ratchasima*: Khao Yai National Park, 14°13'N, 101°11'E, alt. *c.* 800 m, on bark of *Mangifera indica* in evergreen forest, 19.viii.1997, *K. Boonpragob et al.* (RAMK 7958, 8140).

4. Laurera meristosporoides P. M. McCarthy & Vongshewarat, *sp. nov.* Fig. 1
Thallus subepiphloeodalis, pallido flavoviridis, continuus vel leviter rimosus, laevigatus, rugosus aut verrucosus. Pseudostromata prominantia, 1(–4)-carpeae, 0.6–1.2(–2) mm, thallo concoloro. Ascomata fusconigra, immersa. Pseudoparaphyses anastomosantes. Asci 8-spori, 264–316 × 37–55 μm. Ascosporae muriformes, (68–)89(–118) × (18–)21(–24) μm.

Type: Thailand, Phisanulok Province, Phu Hin Rong Kla National Park, 16°59'N, 101°06'E, alt. 1200 m, on bark in montane evergreen forest, 23.ii.1998, *K. Vongshewarat 173* (CANB–holo).

Thallus subepiphloeodal, determinate, continuous to sparingly rimose, smooth, rugose or verrucose, matt to slightly glossy, pale yellowish green, corticate. *Cortex* hyaline, K+ yellowish (thin section), 30–60 μm thick; cells indistinct; walls massively thickened and, apparently, gelatinized; lumina periclinally elongate, sparingly to richly branched. *Photobiont layer* with a discrete outer edge and a diffuse inner edge, 20–30 μm thick. *Photobiont Trentepohlia*; cells 6–16 × 5–12 μm; interstitial hyphae 2–3 μm thick. *Medulla* whitish, merging with the substratum, comprising 2–4 μm wide hyphae that are heavily interspersed with crystals, K–. *Prothallus* not apparent.

Pseudostromata rounded and convex to subglobose or irregular in outline, containing 1(–4) ascomata, 0.6–1.2(–2) mm, concolorous with the thallus apart from the periostiolar area that is plane to slightly convex, pale to dark grey-brown, 0.17–0.42 mm diam., bounded by a whitish ring of exposed medullary material, and with an inconspicuous, central ostiole. *Pseudostromatal wall* an upward extension of

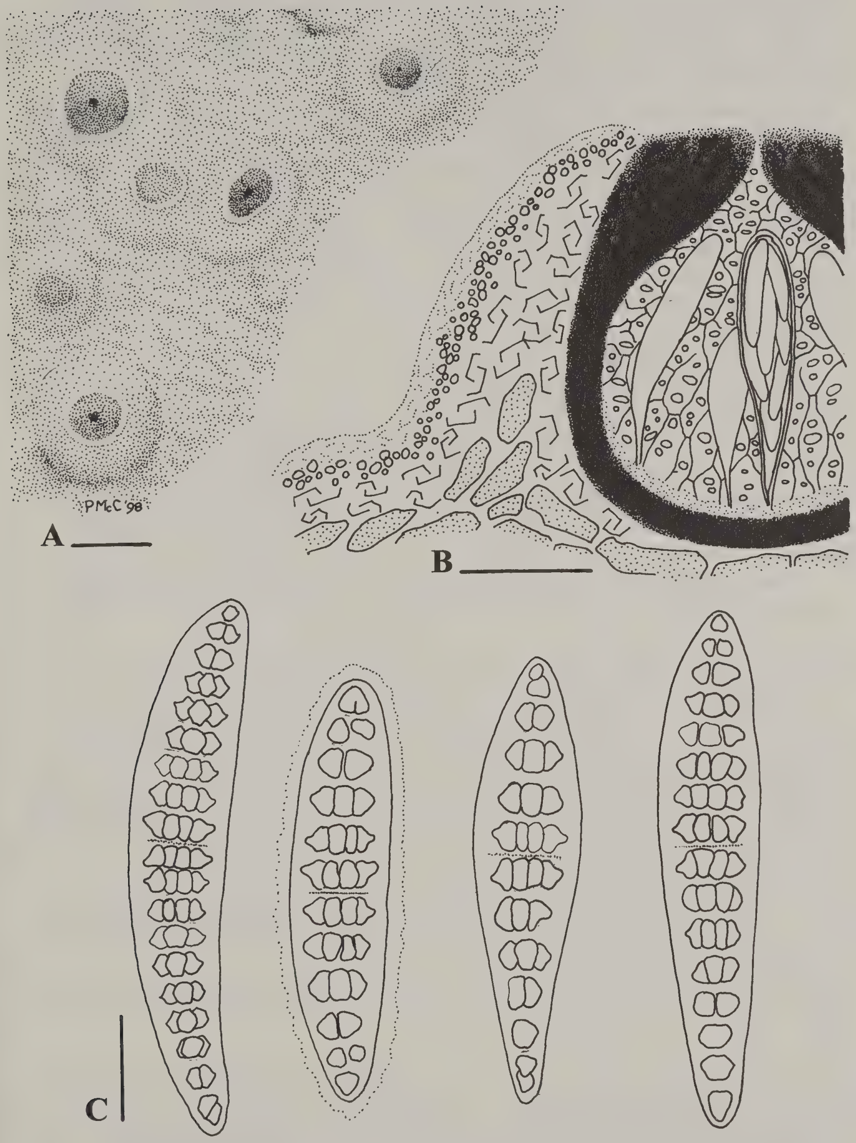

Fig. 1. *Laurera meristosporoides* (holotype). A, Habit of thallus and pseudostromata; B, Part of vertical section of monocarpic pseudostroma and adjacent thallus (semi-schematic); C, Ascospores. Scales A = 0.5 mm, B = 0.1 mm, C = 20 µm.

the thallus. *Ascomata* 0.5–0.9 mm diam. *Excipulum* brown to blackish and 70–110 µm thick near the ostiole, brownish black and 25–40 µm thick at the sides and base. *Hymenium* sparsely to densely inspersed with hyaline lipid globules, Lugol's I–. *Pseudoparaphyses* anastomosing, long-celled, 0.5–0.8 µm thick, growing from the subhymenium and from the excipulum as far up as the ostiole. *Periphyses* absent. *Asci* fissitunicate, 8-spored, cylindroclavate to elongate-clavate, 264–316 × 37–55 µm [*n* = 12]; apex with a broad, convex ocular chamber; stalk long, tapering; ascoplasma Lugol's I+ dark reddish brown. *Ascospores* colourless, muriform, elongate-ellipsoidal, subcylindrical or elongate-fusiform, with (9–)11–15(–18) transverse septa, separating ranks of up to 6 angular or rounded cells, irregularly massed in the ascus, with rounded apices and, sometimes, an uneven, 3–5 µm thick gelatinous sheath, (68–)89(–118) × (18–)21(–24) µm [*n* = 133]; median primary septum remaining distinct; contents clear.

Conidiomata not seen.

Notes: This lichen is characterised by its thin, yellowish green thallus, prominent but comparatively small, mostly monocarpic pseudostromata, with structurally simple walls (basically an upward extension of the thallus), and by the dimensions and degree of septation of its ascospores. Outwardly it is rather similar to the common, eastern Palaeotropical lichen *L. meristospora* (Mont. & Bosch) Zahlbr., but that species has much larger ascospores (170–220 × 32–40 µm) with 33–40 transverse divisions (Letrouit-Galinou 1957).

Laurera meristosporoides is known only from the type locality in montane evergreen forest in north-eastern Thailand near the border with Laos.

5. Laurera phaeomelodes (Müll. Arg.) Zahlbr., *in* A. Engler & K. Prantl, *Nat. Pflanzenfam.* 1, 1: 71 (1903)

Thallus endophloeodal, olive-brown or dark yellowish brown, smooth; photobiont *Trentepohlia*. Ascomata conical to hemispherical to subglobose, black, 0.2–0.5 mm diam., spreading at the base and aggregated into irregular pseudostromata. Hymenium comprising anastomosing pseudoparaphyses and 8-spored, 100–130 × 30–40 µm asci; oily substance absent. Ascospores elongate-ellipsoidal, muriform, with 8–10 transverse divisions, 38–50 × 10–15 µm.

Laurera phaeomelodes has been reported from Cuba and Nigeria (Letrouit-Galinou 1957) and, in the Palaeotropics, from the Andaman Islands in the Bay of Bengal (Makhija & Patwardhan 1988).

SPECIMEN EXAMINED: *Phisanulok*: Phu Hin Rong Kla National Park, 16°59'N, 101°06'E, alt. 1200 m, on bark in montane evergreen forest, 23.ii.1998, *K. Vongshewarat 153, 154* (RAMK).

6. Laurera subdiscreta (Nyl.) Zahlbr., *Cat. Lich. Univ.* 1: 506 (1922)

Thallus endophloeodal, yellowish brown to dark grey-brown, smooth; photobiont *Trentepohlia*. Ascomata solitary in hemispherical to subglobose, dark grey to greenish black, 0.5–0.75 mm diam. pseudostromata. Ascomatal apex concave to plane, 0.1–0.25 mm diam. Hymenium comprising anastomosing pseudoparaphyses and 8-spored, 150–180 × 30–40 µm asci; oily substance absent. Ascospores cylindrical to elongate-ellipsoidal, muriform, with 9–13 transverse divisions, 40–61 × 15–24 µm.

Laurera subdiscreta is known from Bengal, eastern India (Letrouit-Galinou 1957, Makhija & Patwardhan 1988).

SPECIMENS EXAMINED: *Sakonnakhon*: Phu Pan National Park, 16°53'N, 104°25'E, alt. 200 m, on bark in mixed deciduous forest, 7.x.1997, *K. Vongshewarat* (RAMK 8917); *loc. id.*, alt. 400 m, on bark of *Xylia* sp. in dry dipterocarp forest, 24.v.1998, *W. Khumtim 376* (RAMK). *Burirum*: Khao Kadong, 14°50'N, 103°06'E, alt. 300 m, on bark in dry dipterocarp forest, 9.v.1998, *K. Vongshewarat 265* (RAMK).

7. **Polymeridium catapastum** (Nyl.) R. C. Harris, *Acta Amazonica* 14 (Suppl.): 70 (1984) ['1986']

Thallus endophloeodal, pale grey, ecorticate, UV–; photobiont *Trentepohlia*. Perithecia semi-immersed, convex to subconical, black, 0.25–0.4 mm diam.; apex rounded; ostiole apical to eccentric. Pseudoparaphyses anastomosing, interspersed with granular material. Asci 8-spored, broadly to elongate-cylindrical, with a broad ocular chamber. Ascospores colourless, elongate-cylindrical, 3-septate, 22–32 × 7–10 μm, with cylindrical lumina and rounded ends.

Polymeridium catapastum is especially well-known in the Neotropics, also Malaysia, Papua New Guinea, eastern Australia and New Zealand (Harris 1993).

SPECIMEN EXAMINED: *Nakhon Ratchasima*: Khao Yai National Park, Orchid Trail to Hiew Suwat Falls, beside Lam Takhong Creek, 14°13'N, 101°11'E, alt. c. 500 m, on twigs, 4.xi.1997, *P. M. McCarthy T80/97* (CANB, RAMK).

8. **Polymeridium quinqueseptatum** (Nyl.) R. C. Harris, *in* S. C. Tucker & R. C. Harris, *Bryologist* 83: 12 (1980)

Thallus endophloeodal, pale grey, ecorticate, UV–; photobiont *Trentepohlia*. Perithecia semi-immersed to almost superficial, convex, hemispherical or subconical, black, 0.3–0.5 mm diam.; apex usually slightly flattened; ostiole apical. Pseudoparaphyses anastomosing, interspersed with granular material. Asci 8-spored, broadly to elongate-cylindrical, with a broad ocular chamber. Ascospores colourless, elongate-cylindrical, 7-septate, 19–28 × 5–7.5 μm, with cylindrical lumina, rounded ends and a thick gelatinous sheath.

This lichen is already known from the Neotropics, southern Africa, India, north-eastern Australia, the Philippines and Hong Kong (Harris 1993).

SPECIMEN EXAMINED: *Nakhon Ratchasima*: Khao Yai National Park, Orchid Trail to Hiew Suwat Falls, beside Lam Takhong Creek, 14°13'N, 101°11'E, alt. c. 500 m, on twigs, 4.xi.1997, *P. M. McCarthy* T81/97 (CANB, RAMK).

9. **Porina erawanensis** P. M. McCarthy & Vongshewarat, *sp. nov.* Fig. 2

Thallus epiphloeodalis, pallido griseoviridis, adpressus, continuus vel leviter rimosus, rugulosus et verruculosus, 30–60 μm crassus. Perithecia in verrucis thallinis, (0.34–)0.43(–0.55) mm diametro immersa. Centrum 0.18–0.25 mm diametro. Asci 112–140 × 17–24 μm. Ascosporae (11–)15-septatae, elongatae-fusiformes vel elongatae-cylindrici, (48–)58(–72) × (7–)8.5(–9.5) μm.

Type: Thailand, Kanchanaburi Province, Erawan National Park, 14°24'N, 99°09'E, alt. 85 m, on bark in evergreen forest, 22.ix.1997, *K. Vongshewarat* (CANB–holotype; RAMK 8570–isotype).

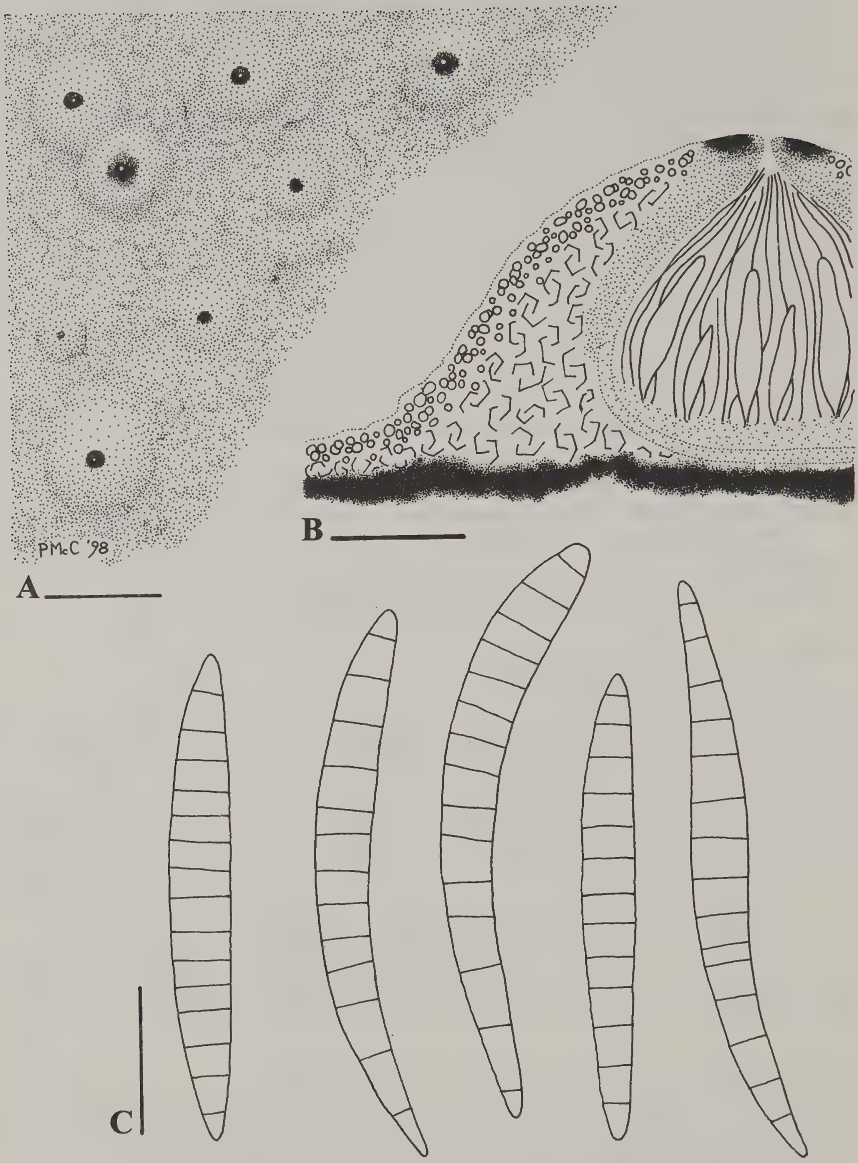

Fig. 2. *Porina erawanensis* (holotype). A, Habit of thallus and perithecia; B, Part of vertical section of perithecium and adjacent thallus (semi-schematic); C, Ascospores. Scales A = 0.5 mm, B = 0.1 mm, C = 20 μm.

Thallus epiphloeodal, determinate, pale greyish green, 30–60 µm thick, closely adpressed to the substratum, continuous to sparingly rimose, matt to slightly glossy, heavily impregnated with crystals. *Surface* rugulose-verruculose, K-, becoming more intensely green when wetted. *Cortex* amorphous to ±prosoplectenchymatous, 5–10 µm thick. *Algae Trentepohlia*, subglobose to globose, 5–10 × 5–9 µm. *Hyphae* 2–3 µm wide. *Prothallus* a blackish line. *Basal layer* brownish black, 20–30 µm thick.

Perithecia very numerous, immersed in thallus-dominated verrucae. *Apex* usually convex, with a dark brown to black, 60–100 µm diam. periostiolar area. *Verrucae* mostly convex, occasionally hemispherical to subconical, (0.34–)0.43(–0.55) mm diam. [*n* = 60]; ostiole pale brown, *c.* 20 µm diam. *Involucrellum* vestigial, apical to dimidiate, brownish black at the apex, orange-brown below (thin section). *Excipulum* medium orange-brown near the ostiole, pale yellowish brown at the base, *c.* 15 µm thick. *Centrum* subglobose to depressed-ovate, 0.18–0.25 mm diam. *Paraphyses* unbranched to sparingly branched, rather stiff, *c.* 1 µm thick. *Periphyses* absent. *Asci* elongate-cylindrical to elongate-obclavate, 8-spored, 112–140 × 17–24 µm [*n* = 15], with a rounded apex containing a chitinoid ring (in Conge red). *Ascospores* colourless, (11–)15-septate, elongate-fusiform to elongate-cylindrical, straight, curved or sigmoidal, irregularly biseriate or mostly massed in the asci, (48–)58(–72) × (7–)8.5(–9.5) µm [*n* = 63], often with a *c.* 1 µm thick gelatinous sheath; contents clear.

Conidiomata not seen.

Notes: *Porina erawanensis* is one of a number of tropical and subtropical species having thallus-dominated perithecial verrucae and elongate, multiseptate ascopores.

In the Neotropics, *P. dolichophora* (Nyl.) Müll. Arg. has perithecial verrucae 0.52–1.1 mm diam., and 13–21-septate ascospores measuring 62–109 × 5–7 µm (McCarthy 1993), while the more commonly saxicolous *P. exserta* Müll. Arg. has 0.35–0.96 mm diam. verrucae and 11–21-septate, 44–70 × 3.5–5.5 µm ascospores (McCarthy 1993). The eastern Palaeotropical and Pacific *P. bellendenica* Müll. Arg. has verrucae of 0.35–0.6 mm diam., each with a broad, blackish periostiolar cap, as well as 9–15-septate ascospores of 32–56 × 3–5 µm (McCarthy 1993). Finally, *P. belanospora* (Nyl.) Müll. Arg., first described from Colombia, and recently reported from India, has rather similar ascospores, but the perithecia are largely immersed and 0.2–0.3 mm diam. (Makhija *et al.* 1994)

This lichen is known only from the type locality in western-central Thailand where it grows in evergreen forest .

10. Porina internigrans (Nyl.) Müll. Arg., *Rep. Australas. Assoc. Advancem. Sci.* 1895: 452 (1895)

This lichen has a thin to rather thick, greyish green thallus, large perithecial verrucae each with a broad, black periostiolar area, and elongate, (7–)9–11(–13)-septate ascospores measuring 51–92 × 9–17 µm. It is already known from the Andaman Islands, Papua New Guinea, north-eastern Australia and New Caledonia (see McCarthy (1994) for a full description and illustrations).

SPECIMEN EXAMINED: *Sukothai*: Ramkhamhaeng National Park, 16°53'N, 99°42'E, alt. 400 m, on bark in dry evergreen forest, 22.x.1997, *K. Vongshewarat* (RAMK 9509.1).

11. Pyrenula anomala (Ach.) Vainio, *Ann. Acad. Sci. Fenn.*, ser. A, 6(7): 189 (1915)
Thallus pale yellowish brown, smooth, corticate, lacking pseudocyphellae, UV–; photobiont *Trentepohlia*. Perithecia numerous, almost superficial, black, 0.25–0.35 mm diam., mostly fused laterally in groups of 5–10(–15); apex plane to concave, 70–150 µm diam.; ostiole apical. Paraphyses simple, not interspersed with granules. Ascospores brown, ellipsoidal, 3-septate, 15–22 × 6–10 µm, uniseriate to irregularly biseriate in the ascus, with a thin endospore and rounded ends.

This pantropical and subtropical species (Harris 1989) is known, in the eastern Palaeotropics, from India, Papua New Guinea and the Philippines.

SPECIMENS EXAMINED: *Chanthaburi*: Numtokphliu National Park, 12°29'N, 102°12'E, alt. 85 m, on bark in evergreen forest, 18.ix.1997, *K. Vongshewarat* (RAMK 8743). *Chiang Mai*: Queen Sirikit Botanic Garden, 18°48'N, 98°58'E, on bark of *Helicia* sp., *K. Vongshewarat 122* (RAMK). *Lumpang*: Khuntan, 18°25'N, 99°16'E, alt. 1100 m, on bark in dry evergreen forest, 6.iii.1998, *K. Vongshewarat 62* (RAMK). *Sakonnakhon*: Phu Pan National Park, 16°53'N, 104°25'E, alt. 345 m, on bark in dry evergreen forest, 7.x.1997, *K. Vongshewarat* (RAMK 8414). *Burirum*: Khao Kadong, 14°50'N, 103°06'E, alt. 300 m, on bark of *Buchanania* sp. in dry dipterocarp forest, 8.v.1998, *K. Vongshewarat 250* (RAMK).

12. Pyrenula macularis (Zahlbr.) R. C. Harris, *Mem. New York Bot. Gard.* 49: 94 (1989)
Thallus epiphloeodal, medium yellowish brown, smooth, corticate, with white pseudocyphellae, UV–; photobiont *Trentepohlia*. Perithecia almost completely immersed, 0.22–0.32 mm diam.; involucrellum thin. Paraphyses simple, not interspersed with granules. Ascospores pale brown, submuriform, with 5–7 transverse divisions separating rows of 2–4 locules, 30–45 × 12–19 µm, with a thick endospore, uniseriate to biseriate in the ascus; postmature ascospores with a reddish oily substance.

The distribution of this pantropical lowland species (Harris 1989) includes Papua New Guinea and northern Australia.

SPECIMEN EXAMINED: *Rayong*: 12°38'N, 101°27'E, on bark of *Rhizophora macronata* in mangrove forest, 2.v.1998, *K. Vongshewarat 323* (RAMK).

13. Pyrenula parvinuclea (Meyen & Flot.) Aptroot, *in* A. Aptroot *et al.*, *Biblioth. Lichenol.* 64: 152 (1997)
Thallus subepiphloeodal, white to pale grey, rimose, uneven, ecorticate, UV–; photobiont *Trentepohlia*. Perithecia numerous, semi-immersed to superficial, hemispherical to subglobose, dark brown to black, 0.2–0.37 mm diam.; apex rounded; ostiole apical, inconspicuous or in a shallow depression. Paraphyses simple, not interspersed with granules. Ascospores pale brown, elongate-ellipsoidal, uniseriate, submuriform, with 4 ranks of 2–4 locules each, 10–20 × 7–10 µm.

This uncommon, lowland Palaeotropical lichen has recently been reported from Papua New Guinea (Aptroot *et al.* 1997).

SPECIMENS EXAMINED: *Samotprakhang*: Muang Boran, 13°36'N, 100°36'E, on bark of isolated tree, 11.xi.1997, *P. M. McCarthy T90/97* (CANB). *Chachesoa*: 13°36'N, 100°37'E, on bark of *Ceriops tagal* in mangrove forest, 14.iii.1998, *K. Vongshewarat 204* (RAMK).

14. Pyrenula subferruginea (Malme) R. C. Harris, *Mem. New York Bot. Gard.* 49: 101 (1989)

Thallus epiphloeodal, rather thick, pale greyish green, continuous, uneven; surface with clusters of white crystals, UV–; photobiont *Trentepohlia*. Perithecia solitary, inconspicuous, blackish, 0.5–0.75 mm diam., almost completely overgrown by the thallus, or with an exposed, blackish, 0.2–0.5 mm diam. periostiolar area; ostiole eccentric to lateral, at first concolorous with the thallus, becoming dark grey-brown. Paraphyses simple, not interspersed with granules. Ascospores uniseriate in the ascus, broadly to elongate-cylindrical or citriform, medium to dark brown, 3-septate, 18–27 × 8–12 µm; median cells rounded or angular; apical cells elongate and pressing against the end walls.

This lichen is known from several localities in Brazil and U.S.A. (Harris 1989) and from Papua New Guinea (Aptroot *et al.* 1997).

SPECIMEN EXAMINED: *Kanchanaburi*: Erawan National Park, 14°24'N, 99°09'E, alt. 85 m, on bark in evergreen forest, 22.ix.1997, *K. Vongshewarat* (RAMK 8576).

15. Trypethelium nigroporum Makhija & Patw., *J. Hattori Bot. Lab.* 73: 202 (1993)

Thallus epiphloeodal, grey-green to yellowish green, smooth, thickly corticate; photobiont *Trentepohlia*. Pseudostromata very numerous, mostly solitary or paired, otherwise in rows or clusters of up to 8, hemispherical to subglobose, 0.25–0.5 mm diam., externally dark brown to black, K+ reddish, internally partly carbonised, partly orange-brown; apex plane to slightly convex, with a distinct rim, 0.1–0.35 mm diam. and an inconspicuous ostiole. Pseudoparaphyses anastomosing, not interspersed with oil globules. Excipulum dark brown to black. Asci 8-spored, with a broad ocular chamber. Ascospores colourless, 4–7-septate, 30–40 × 7–10 µm, with cylindrical lumina.

This species was recently described from Nagaland and Meghalaya in India (Makhija & Patwardhan 1993).

SPECIMENS EXAMINED: *Loei*: Naheaw, 17°05'N, 101°05'E, alt. 750 m, on bark of *Bombax anceps* in evergreen forest, 8.xi.1995, *P. Mongkolsuk* (RAMK 5852). *Sakonnakhon*: Phu Pan National Park, 16°53'N, 104°25'E, alt. 200 m, on bark in mixed deciduous forest, 7.x.1997, *K. Vongshewarat* (RAMK 8920.3).

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MOLECULAR TAXONOMY OF CERIPORIA

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ABSTRACT

Mitochondrial small subunit ribosomal DNA sequences of *Ceriporia* and its related genera were determined to verify present taxonomic systems in terms of phylogenetic relationships at generic level. *Ceriporia* proved to be monophyletic. *Ceriporia viridans* had no phylogenetic relationship to *Phlebia radiata*. *Ceriporia* species and *Oxyporus* formed a monophyletic clade. *Ceriporia-Oxyporus* and *Phanerochaete-Pulcherricium-Bjerkandera* clades were too insignificantly grouped to become a monophyletic group. The presence or absence of clamps on generative hyphae has a good taxonomic and phylogenetic significance to separate *Ceriporia* and *Ceriporiopsis*. *Ceriporia viridans* has diverged from other *Ceriporia* species earliest in the genus and has developed ceriporiones as an autapomorphic character.

Key words: *Ceriporia*, *Ceriporiopsis*, *Oxyporus*, *Phlebia*, phylogeny

INTRODUCTION

Ceriporia was proposed by Donk with *C. viridans* as a type species from Bourdot & Galzin's *Poria* sect. *Chrooporae* group which was an unnatural assemblage of resupinate polypores (Donk, 1967; Donk, 1968). It originally contained species with monomitic hyphal systems of simple-septate to clamped generative hyphae and white rot activity. *Ceriporiopsis* Domanski was later segregated for the species lacking cupular nidulant young pores and having clamps in generative hyphae (Donk, 1967).

In *Ceriporia,* 11 species have been described in North America and Europe (Gilbertson and Ryvarden, 1986; Ryvarden and Gilbertson, 1993). Generic characters defining the species included in *Ceriporia* are resupinate and soft basidiocarps of white, pink, orange or green pore surfaces, monomitic hyphal systems of simple-septate generative hyphae (except for *C.*

excelsa and *C. metamorphosa* with rarely present clamps), no cystidia or other sterile hymenial elements, cylindric to allantoid and hyaline basidiospores with negative reaction in Melzer's reagent, and white rot activity on dead conifers and hardwoods (Ryvarden and Gilbertson, 1993).

There has been some doubt on the monophyly of *Ceriporia* based on chemotaxonomic and other related data (Ryvarden, 1991). *Ceriporia viridans,* the type species of the genus, produces ceriporiones which give a reddish reaction with acetone and impart a cherry red color to the acetone extract, but no other species in the genus have been reported to have such compounds (Ryvarden, 1991). However, *Merulius tremellosus* of the Meruliaceae and *Phlebia radiata* of the Corticiaceae are known to have compounds known as merulinic acids closely related to ceriporiones (Gill and Steglich, 1987). Moreover, above-mentioned three species have identical spores which are hyaline, inamyloid, allantoid to cylindric, and (3.5-) 4-5 (-6) x (1-) 1.5-2 μm in dimension (Gilbertson and Ryvarden, 1986; Breitenbach and Kränzlin, 1986; Eriksson *et al*., 1981). Based on the similarity of chemical compounds and spore morphology, *Ceriporia viridans* was sometimes assumed to be phylogenetically related to *Merulius tremellosus* and *Phlebia radiata* (Ryvarden, 1991). The latter two species also have resupinate basidiocarps and monomitic hyphal systems although there are clamps on their generative hyphae.

In cultural characters of *Ceriporia* species, some variations occur again. *Ceriporia reticulata* is reported to have partially thick-walled, enlarged, short-celled and simple-septate old hyphae, but *Ceriporia excelsa* is described to show a positive gum guaiac test and has simple-septate hyphae, but with rare single or double clamps, encrusted with octahedral crystals (Lombard and Gilbertson, 1966). *Ceriporia purpurea* is reported to develop a negative reaction for the gum guaiac test with no diffusion zone on gallic acid agar or tannic acid agar and has simple-septate hyphae sometimes heavily encrusted with yellow crystals (Nakasone and Gilbertson, 1978).

Suggestions on relationships of *Ceriporia* to other genera have been given by some mycologists. Ryvarden (1991) grouped *Ceriporia* with *Flabellophora, Heterobasidion, Hydnopolyporus, Henningsia, Leucophellinus, Macrohyporia, Meripilus, Oxyporus, Rigidoporus* and *Physisporinus* together in the *Rigidoporus* group which has monomitic to dimitic hyphal systems, simple-septate generative hyphae, and white rot habit. In this group, *Ceriporia* deviates from other genera in spore morphology by having allantoid to cylindric spores. According to Ryvarden (1991), when the clamp is neglected, *Ceriporia* and *Ceriporiopsis* could be merged into a single genus. Jülich (1981) treated *Ceriporia*, with *Rigidoporus* and *Oxyporus* together, in the family Rigidoporaceae of his new order Phanerochaetales. The relatedness of *Oxyporus* and *Rigidoporus* has been pointed out by several authors (Pouzar, 1966; Ryvarden and Gilbertson, 1994). *Oxyporus* and *Rigidoporus* share common characters like monomitic hyphal systems of simple-septate hyphae, white rot, more or less globose spores and presence of cystidia. But *Ceriporia* differs from these two genera by having cylindric spores and no sterile elements.

Taxonomic studies in fungal groups recently have been done using more and more molecular data. Hibbett and Donoghue (1995) conducted molecular phylogenetic studies on the Polyporaceae using mitochondrial small subunit ribosomal DNA (mt ssu rDNA) sequences, where *C. purpurea* and *C. viridans* were included. The present study was accomplished to test the monophyly of *Ceriporia* and to see possible relationships between *C. viridans* and *Phlebia radiata* and phylogenetic relationships of *Ceriporia* to other genera of the families Polyporaceae and Corticiaceae, using mt ssu rDNA regions as molecular characters to answer present taxonomic questions.

MATERIALS AND METHODS

Culture of fungi and DNA extraction

Strains used for this study are listed in Table 1. Three *Ceriporia* species, two *Ceriporiopsis* species and *Oxyporus latemarginata* have been sequenced and their sources and sequence accession numbers are included in Table 1. *Meripilus giganteus* and *Heterobasidion annosum* have been chosen according to the Ryvarden's scheme that both are included in the *Rigidoporus* group based on monomitic to dimitic hyphal systems with simple septa and white rot activity (Ryvarden, 1991). *Auricularia auricula* was chosen as an outgroup, based on the work of Swann and Taylor (1993), for rooting phylogenetic trees.

Cultures were performed on ME broth (malt extract 2%, peptone 0.5%) at 24℃, harvested, lyophilized and ground to fine powder. DNA extraction was performed after Bruns *et al.* (1990). Samples of 25-30 mg were placed in 1.5 ml microcentrifuge tubes, suspended in 700 μl of 50 mM EDTA, 50 mM Tris of pH 7.5, and 3% SDS, and then incubated for 30-60 minutes at 65℃. DNA was extracted once with phenol/chloroform 1:1, once with chloroform, and then precipitated by addition of 50 μl of 3 M ammonium acetate, followed by 500 μl of isopropanol, washed once with 70% ethanol, and resuspended in 250 μl of TE buffer. Extracted DNA was stored at -20℃ for further study.

PCR, cloning and sequencing

Mt ssu rDNA gene was amplified from extracted total genomic DNA using MS1 and MS2 primers (White *et al.*, 1990). PCR amplification of DNA was performed by *Taq* DNA polymerase in 10 mM Tris-HCl (pH 9.0 at 25℃), 50 mM KCl, 0.1% Triton X-100, 2 mM $MgSO_4$, 0.2 mM of each dNTP, and 0.5 μM of each primer with about 0.2 μg template DNA, diluted 100-fold by DDW, and 1 unit of *Taq* DNA polymerase per PCR reaction. Total volume was adjusted at 25 μl and PCR reactions were performed in a

Table 1. Fungal species, family ranks, and GenBank accession numbers of mitochondrial small subunit rRNA sequences of analyzed taxa.

Species	Family	Source	GenBank Accession
Auricularia auricula-judae (L.: Fr.) J. Schroet.[1]	Auriculariaceae		U27022
Bjerkandera adusta (Willd.: Fr.) Karst.	Polyporaceae		U27025
Ceriporia excelsa (Lund.) Parm.	Polyporaceae	CBS 509.85	AF041090
Ceriporia purpurea (Fr.) Donk[2]	Polyporaceae	CBS 343.63	AF041091
Ceriporia purpurea (Fr.) Donk[3]	Polyporaceae		U27029
Ceriporia reticulata (Pers.: Fr.) Dom.	Polyporaceae	CBS 461.50	AF041095
Ceriporia viridans. (Berk. et Br.) Donk.	Polyporaceae		U27030
Ceriporiopsis rivulosa (Berk. et Curt.) Gilbn. et Ryv.	Polyporaceae	CBS 434.48	AF041092
Ceriporiopsis resinascens var. *pseudogilvescens* (Rom.) Dom.	Polyporaceae	CBS 525.92	AF041093
Heterobasidion annosum (Fr.) Bref.	Polyporaceae		U27042
Meripilus giganteus (Fr.) Karst.	Polyporaceae		U27053
Oxyporus latemarginata (E. J. Durand et Mont.) Donk	Polyporaceae	ATCC 9408	AF041094
Oxyporus sp.[4]	Polyporaceae		U27054
Phanerochaete chrysosporium Burds.	Corticiaceae		U27063
Phlebia radiata Fr.	Corticiaceae		AF026649
Pulcherricium caeruleum (Fr.) Parm.	Corticiaceae		U27057

[1] Instead of this name, *Auricularia auricula* was used in text and Fig. 1.

[2] This strain was indicated as *Ceriporia purpurea* 1 in Fig. 1.

[3] This strain was indicated as *Ceriporia purpurea* 2 in Fig. 1.

[4] This strain was identified and sequenced by Hibbett and Donoghue (1995).

programmable thermal controller with the following parameters: an initial extension step for 3 min at 94℃, 30 s at 94℃ for denaturation, 30 s at 55℃ for annealing, and 1 min at 72℃ for extension, which processes were cycled 30 times, and then final extension for 30 min at 72℃ was followed.

PCR products were observed by electrophoresing 3 μl from each tube on 1.5% agarose gel containing EtBr in Tris-acetate EDTA (TAE) buffer. The presence of a single bright band in each lane was a check for a successful amplification. Negative control which contains all components except DNA template was always included to detect any possible contamination during PCR reaction. Each PCR reaction tube was added with 150 μl of sterilized DDW and extracted once with equal volume of chloroform. And 0.1 volume of 3 M sodium acetate and 2.5 volume of ethanol were added, precipitated by centrifugation for 15 min at 12,000 rpm at 4℃, washed once with 70% ethanol, and evaporated using vacuum evaporator. Purified PCR products were cloned using the Promega T-vector system. The ligation conditions are as follows: 25 ng of pGEM T-vector, 100 ng of PCR products and 3 Weiss unit of T4 ligase in 10 μl of reaction mixture. Ligation reaction was performed at 14℃ overnight. Ligation mixture was transformed according to the method of Sambrook *et al.* (1989). Cloned plasmids were denatured by alkaline method (0.2 M NaOH, 0.2 mM EDTA, at 37℃ for 30 min) and were used for DNA sequencing. The dideoxy chain termination method (Sanger *et al.*, 1977) was used for manual DNA sequencing using Sequenase II 2.0 kit (Amersham) according to the manufacturer's manual.

Sequence data analyses

Sequences were aligned using CLUSTAL W (Thompson *et al.*, 1994) and then visually optimized to allow maximal alignment. To analyze data, most parsimonious trees were found for data sets using PAUP 3.1.1. (Swofford, 1993). Gaps were treated as missing data and options were limited to heuristic searches with simple addition sequence, TBR swapping, MAXTREES unrestricted and MULPARS on. To evaluate the strength of support for branches of parsimonious trees, 100 replications of bootstrap analysis (simple addition sequence, TBR swapping, MAXTREES 10) were performed. For a statistical comparison of the fit of the data to hypothetical trees, likelihood variance test (Kishino and Hasegawa, 1989) was performed using DNAMLK (maximum likelihood with molecular clock) of PHYLIP 3.5c (Felsenstein, 1993).

RESULTS AND DISCUSSIONS

DNAs amplified by PCR using MS1 and MS2 primers were about 521 to 700 bp long in size. From 845 sites aligned by CLUSTAL W, about 50 bases from both 5′ and 3′ ends and unalignable internal sites were excluded. Parsimony analysis of the data set produced 4 equally parsimonious trees of 615 steps. A strict consensus (Fig. 1) tree was generated from four equally

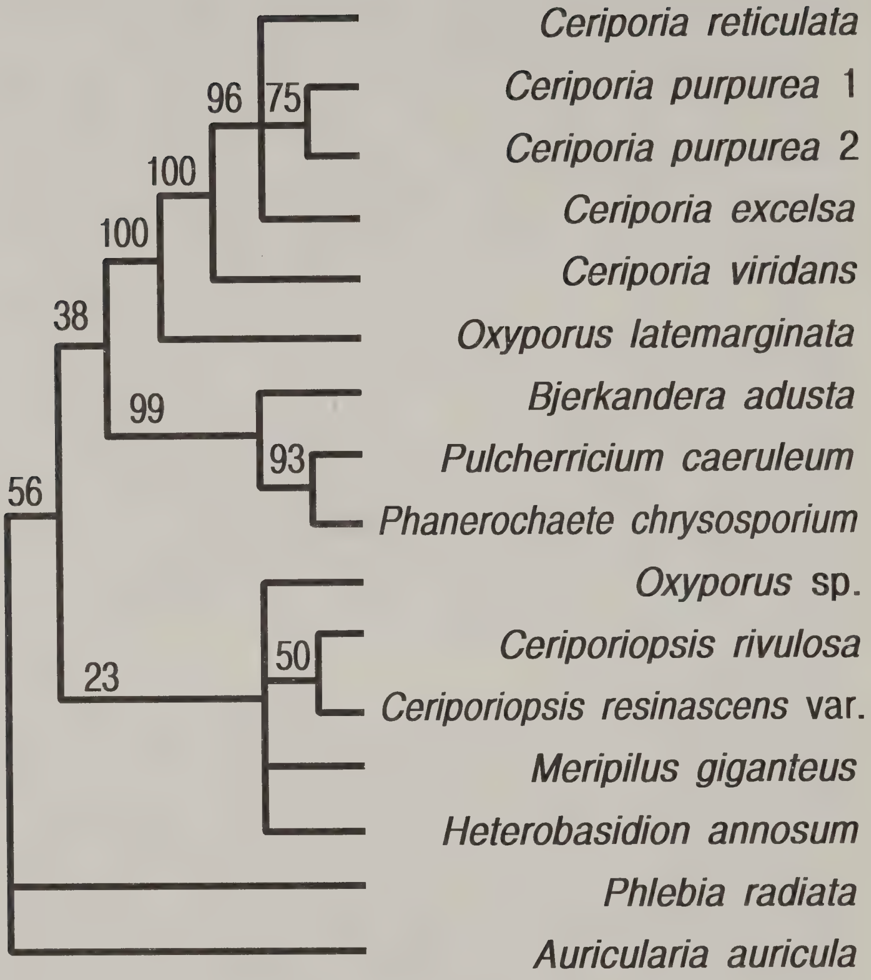
Ceriporia reticulata
Ceriporia purpurea 1
Ceriporia purpurea 2
Ceriporia excelsa
Ceriporia viridans
Oxyporus latemarginata
Bjerkandera adusta
Pulcherricium caeruleum
Phanerochaete chrysosporium
Oxyporus sp.
Ceriporiopsis rivulosa
Ceriporiopsis resinascens var.
Meripilus giganteus
Heterobasidion annosum
Phlebia radiata
Auricularia auricula
96
75
100
100
38
99
93
56
50
23

Fig. 1. Phylogenetic tree of *Ceriporia* and its compared polyporoid and corticioid taxa based on mt ssu rDNA sequences. This is a strict consensus tree generated from four equally parsimonious trees of 615 steps. Bootstrap values computed from 100 replications are given on appropriate branches. *Auricularia auricula* was used as an outgroup. *Ceriporiopsis resinascens* var. corresponds to *Ceriporiopsis resinascens* var. *pseudogilvescens*. Consistency index (CI) = 0.673, homoplasy index (HI) = 0.327, retention index (RI) = 0.631, rescaled consistency index (RC) = 0.425.

parsimonious trees, and statistical supports by 100 bootstrap replications were shown on branches of the tree.

In the tree of Fig. 1, *Ceriporia* taxa formed a completely monophyletic lineage supported by 100% bootstrap value. *Oxyporus latemarginata* grouped with the *Ceriporia* lineage by absolute bootstrap support of 100%. The clade for *Bjerkandera adusta, Pulcherricium caeruleum* and *Phanerochaete chrysosporium* made a group with the one for *Ceriporia* and *O. latemarginata* but very weakly. The other *Oxyporus* species grouped with another clades. Two *Ceriporiopsis* species grouped together by moderate bootstrap support of 50%. But *Phlebia radiata* didn't group with any clade of the tree.

The statistical test, to see if *Ceriporia viridans* is phylogenetically related to *Phlebia radiata* or not, showed that the testing hypothesis was highly unlikely (652 steps, log likelihood = −3865.68) and its tree length was 37 steps longer than that of the most parsimonious tree (615 steps, log likelihood = −3793.68). Another hypothesis which relates *Ceriporia* to the *Ceriporiopsis* group was also highly unlikely (646 steps, log likelihood = −3947.55) compared to the most parsimonious tree. Results of DNAML analyses showed that hypothetical trees were significantly worse than the most parsimonious tree. However, the present results were incongruent with Ryvarden's concepts that *Ceriporia viridans* may be phylogenetically related to the *Phlebia* group of the Corticiaceae and that some species of *Ceriporia* may have originated from corticioid species while others may represent reduced lines of polypores (Ryvarden, 1991).

The separation of *Ceriporia* and *Ceriporiopsis* into distinct genera based on the presence or absence of clamps was strongly supported by the present analysis. As emphasized by many authors (Donk, 1971; Teixeira, 1962; Furtado, 1966; Ryvarden, 1991), the septation of generative hyphae is quite important in the taxonomy of the Polyporaceae and the current taxonomic system of the Polyporaceae is justifiable in this respect. The unrelatedness of the clade for *Ceriporia* and *Oxyporus* to the one for *Phanerochaete, Pulcherricium* and *Bjerkandera* can be found in different septations of generative hyphae. Both *Ceriporia* and *Oxyporus* have simple-septate hyphae, while *Phanerochaete, Pulcherricium* and *Bjerkandera* have constant or occasional clamps on generative hyphae. But, the two groups are quite similar in that they have monomitic hyphal systems, thin-walled, hyaline, non-amyloid spores, and white rot activity on attacked wood.

Two *Oxyporus* species, *O. latemarginata* sequenced by authors and *O.* sp. sequenced by Hibbett and Donoghue (1995), were separated into two distinct clades and showed quite a dissimilarity to each other. In the analysis of Hibbett and Donoghue (1995), this *Oxyporus* sp. was connected to the clade for *Trichaptum, Inonotus* and *Phellinus*, but no phylogenetic considerations could be found in their discussions. In current taxonomy of the Polyporaceae, the genus *Oxyporus* is regarded much related to the genus *Rigidoporus* and *Ceriporia* (Pouzar, 1966; Jülich, 1981; Ryvarden, 1991). It is thus believed that the linking of *Oxyporus* to *Ceriporia* can be more likely than the linking of *Oxyporus* to the clade for *Trichaptum, Inonotus* and *Phellinus*. This fact was also supported by the analysis done with 18S rDNA sequences by authors (unpublished data).

No strong bootstrap support was placed on the relationship of the clade for *Ceriporia* species and *O. latemarginata* with the one for *Ceriporiopsis* species, *Meripilus giganteus* and *Heterobasidion annosum*. Hibbett and Donoghue (1995) once linked *Heterobasidion annosum* to the clade for *Stereum, Lentinellus, Auriscalpium* and *Bondarzewia* which share the amyloidity of basidiospores, without appropriate remarks in their discussions. For the phylogenetic position of *Meripilus giganteus*, little can be said at present. *Meripilus giganteus* has been suggested to be related to the genus *Grifola* based on similar basidiocarp structures but differs only in the septation of generative hyphae (Jülich, 1981; Ryvarden, 1991; Pouzar, 1966). Future molecular works on *Grifola* would reveal the proposed relationship of *Meripilus* and *Grifola.*

As once suggested by Jülich (1981) and Ryvarden (1991), *Ceriporia* and *Oxyporus* proved to be phylogenetically related. Both genera have annual, resupinate to effused-reflexed, soft, easily separable basidiocarps, monomitic hyphal system with simple-septate generative hyphae with no special type, thin-walled, inamyloid, and hyaline spores and white rot. Different characters can be found in pore surface colors. *Oxyporus latemarginata* is white, while *Ceriporia* species is more or less red or pinkish on pore surfaces. Pore shape also differs in that *O. latemarginata* has angular pores while *Ceriporia* species have circular ones. The most important character separating these two genera is cystidia. No cystidia or sterile elements are seen in *Ceriporia* except for hyphoid elements in *C. purpurea*, but *O. latemarginata* have apically encrusted cystidia (Ryvarden and Gilbertson, 1993; Ryvarden and Gilbertson, 1994).

In *Ceriporia*, *C. viridans* might have branched earlier from other *Ceriporia* species. And this coincides with the fact that insertion sequences are present in mt ssu rDNA sequences in most *Ceriporia* species. While there is no insertion sequence present in *C. viridans,* about 50 bp insertion sequence occurs in *C. excelsa* and about 100 bp insertion sequences in *C. reticulata* and *C. purpurea*. And only *C. viridans* has been reported to produce ceriporiones (Ryvarden, 1991). As no other *Ceriporia* species have been known to make such compounds, this character seems to have arisen only in *C. viridans* after it has diverged from other *Ceriporia* species. Another character which is compatible with the phylogenetic tree is the color of the tube. *Ceriporia viridans* has a white tube layer, but *C. purpurea, C. reticulata* and *C. excelsa* have white to pinkish tube and subiculum. The pinkish color of tube and subiculum seems to be a synapomorphic character acquired by *Ceriporia* species other than *C. viridans* after they have diverged from *C. viridans.*

The branching orders in *C. purpurea, C. reticulata* and *C. excelsa* are uncertain and, at present, the three species are treated as polytomies. Each species has its own characters. *C. reticulata* has larger spores than the rest, *C. excelsa* has single or multiple clamps in culture, and *C. purpurea* has hyphoid sterile elements. The low sequence divergence between *Ceriporia* species suggests that above-listed characters are only useful at subgeneric levels (Gilbertson and Ryvarden, 1986; Ryvarden and Gilbertson, 1993).

Ceriporia is apparently monophyletic at least for currently investigated strains and is closely related to *Oxyporus*. On the contrary, *Ceriporia* is not actually related to *Ceriporiopsis*, which proves the importance of presence or absence of clamp connections in defining genera of the Polyporaceae. For the present, it is recommended that more sufficient molecular data derived from other divergent genes or spacer regions need to be added and compared together for consolidated conclusions on taxonomic systems and phylogenetic relationships of *Ceriporia* and its related taxa.

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SEPTORIA HODGESII SP. NOV.: A POTENTIAL BIOCONTROL AGENT FOR MYRICA FAYA IN HAWAIʻI

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Abstract

Septoria hodgesii sp. nov. is described. This fungus is a common leaf pathogen of *Myrica cerifera* in the southeastern U. S., where it usually has been identified as *S. myricae*. It also has been shown by artificial inoculation to be pathogenic on *M. faya*, an introduced forest weed in Hawaiʻi. Comparison of *S. hodgesii* with the types of *S. myricae*, from *M. cerifera* and *S. myricata*, from *M. gale*, shows it to be distinct from both species.

Keywords: *Myrica* spp., *Septoria myricae*, *Septoria myricata*

Myrica faya Ait., commonly known as fayatree, was introduced to Hawaiʻi in the late 1800s by Portuguese immigrants from the Azores and/or Madeira. Subsequently, this species has become a noxious weed in mid to upper elevation forests, pasture, and rangeland. In efforts at biological control, several pathogenic fungi and insects have been found in its native habitats and evaluated for their effectiveness in Hawaiʻi (Lutzow-Felling *et al.*, 1995). In addition, a fungus commonly identified as *Septoria myricae* Ell. & Everh., which causes a leaf spot on wax myrtle *(M. cerifera* L.) in the southeastern U. S. (Schubert, 1986) (Fig. 1), has been tested and shows promise as a biocontrol agent for *M. faya* (Gardner *et al.*, in prep.).

The *Septoria* sp. used in the above-mentioned biological control studies was received from Dr. Charles S. Hodges, Department of Plant Pathology, North Carolina State University, and compared with the type of *S. myricae* and found not to be conspecific. The same results were obtained from comparison with the type of *S. myricata* Dear. & House, reported from *M. gale* L. No other species of *Septoria* have been reported from *Myrica* spp. and the fungus is described as new.

Septoria hodgesii D. Gardner, sp. nov. Figs. 1, 4, 5

Maculae mortuae 3–6 mm diameter, amphigenae, coalescentes ut maculas mortuas forment. Conidiomata pycnidialia hypophyllae, per texturam mortuam dispersa, sphaerica usque ad ampuliformia, 40–60 µm diametro, valde ostiolata. Conidia filiformia, recta aut curvata, (20–)30–90(–110) × 2–5 µm, 3–7 septate.

In foliis vivis *Myrica* spp.

Holotypus BPI 745886; Hawai'i: Hawai'i Volcanoes National Park, Crater Rim Trail.
Isotypus BISH 652531

Necrotic spots 3–6 mm in diameter, amphigenous, coalescing to form necrotic blotches, pycnidial conidiomata hypophyllous, scattered throughout necrotic tissue, spherical to flask-shaped 40–60 µm in diameter. Conidia filiform, straight, curved or sinuous, (20–)30–90(–110) × 2–5 µm, 3–7 septate.

Holotype BPI 745886, Hawai'i: Hawai'i Volcanoes National Park, Crater Rim Trail
Isotype BISH 652531

Etymology: Named for Dr. Charles S. Hodges, Jr.

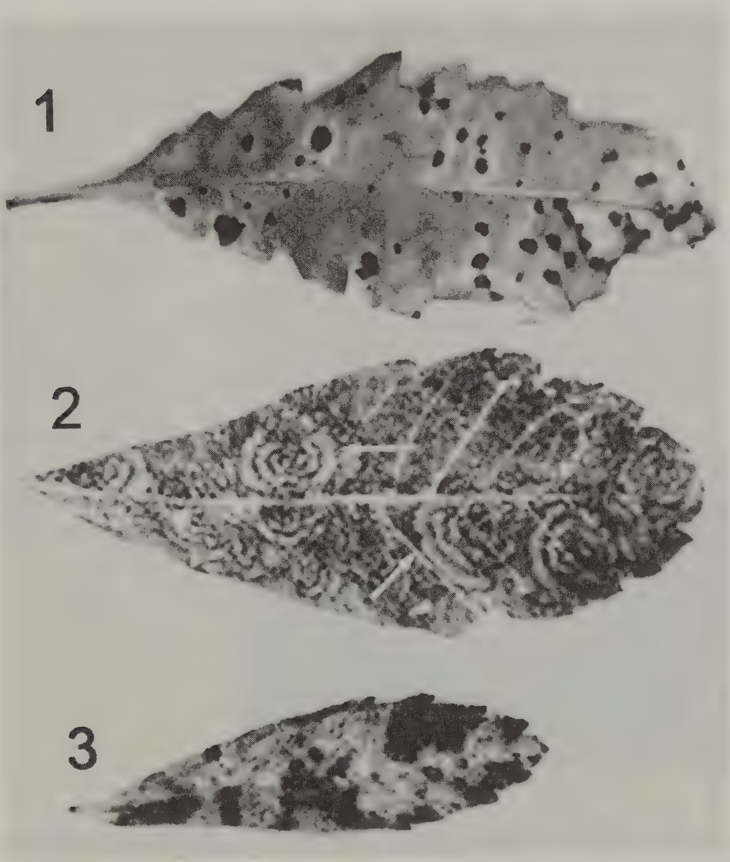
Key to species of *Septoria* on *Myrica*:

1. Conidia bacillus-like, less than 13 µm long; leaf spots concentric rings . . . . . . *S. myricae*
1. Conidia longer than 14 µm 2
    2. Conidia 15–33 µm long; leaf spots indistinct blotches . . . *S. myricata*
    2. Conidia 30–110 µm long; leaf spots distinctly necrotic . . *S. hodgesii*

Specimens of *S. myricae* examined: BPI US0382102, type on *Myrica cerifera*, Newfield, N. J., Sept. 1893, J. B. Ellis, (notation on packet: "isotype? Sept. 1895 given as date of type collection"); BPI US0382104, on *M. cerifera*, Newfield, N. J., Sept. 1893, J. B. Ellis & B. M. Everhart, (notation on packet: "isotype ?"); BPI US0382103, on *M. cerifera*, Newfield,

N. J., Sept. 1895, J. B. Ellis & B. M. Everhart, (notation on packet: "isotype"); BPI US0522077, on *M. cerifera*, Newfield, N. J., Sept. 1895, J. B. Ellis & B. M. Everhart.

Specimens of *S. myricata* examined: NYS (H. D. House 387), type on *M. gale*, Long Lake, Hamilton Co., N. Y., 19 Sept. 1924, H. D. House; DAOM (J. Dearness 5823), on *M. gale*, Long Lake, Hamilton Co., N. Y., 19 Sept. 1924, H. D. House, (notation on packet: "no other *Septoria* on *Myrica* or *Comptonia*").



Figs. 1–3. *Myrica* spp. leaf spots caused by species of *Septoria*. 1. *Myrica faya* with leaf spots of *S. hodgesii*. 2. Type 1893 specimen of *M. cerifera* infected with *S. myricae*. Note the concentric leaf spots (arrows). 3. A 1924 herbarium specimen of *M. gale* infected with *S. myricata*.

My examination of infected leaves of original *S. myricae* collections confirmed Ellis and Everhart's (1897) description of the spots as "often interruptedly confluent over the entire leaf, giving it a mottled appearance; spots sometimes subconcentrically marked." I found much of the upper and lower leaf surfaces covered with large, circular or circuitous lesions comprised of concentric, often coalescing rings creating an intricate pattern (Fig. 2). The hypophyllous fruiting bodies from the 1893 and 1895 collections were grouped in clusters and were mostly open and empty, the conidia having been expelled. Few fertile conidiomata were located. The conidia found were consistent in general with those described for this species, although some measured somewhat longer than the 8–10 μm range given (Fig. 6). Leaf spots caused by *S. hodgesii* have sharply delimited margins with necrotic centers (Fig. 1). Coalescence of individual lesions in heavily infected tissue results in larger necrotic areas.

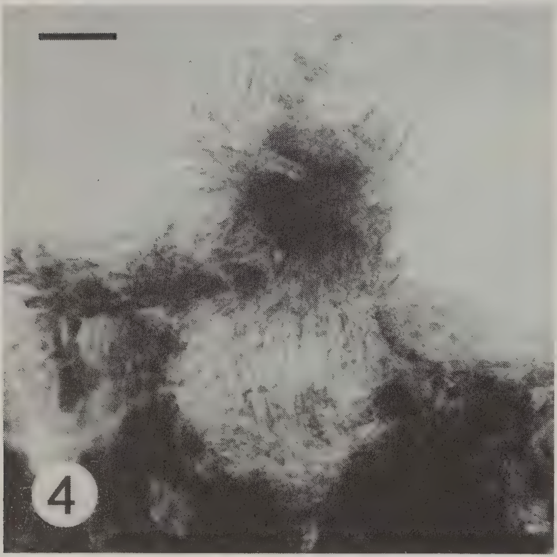
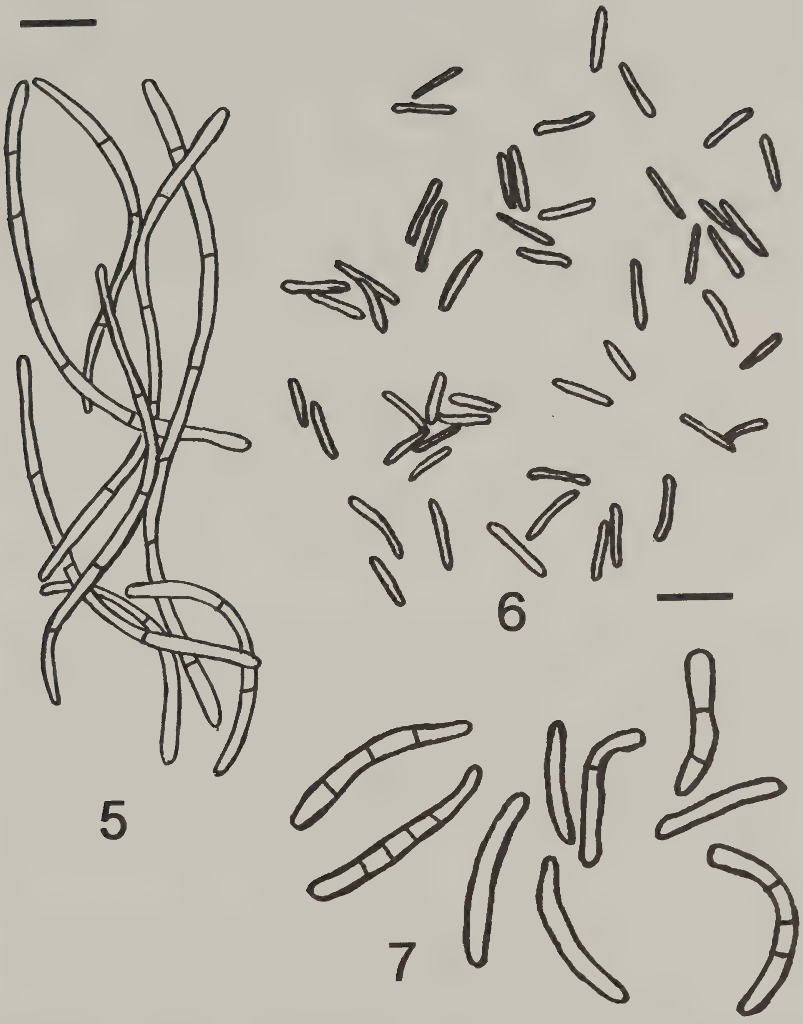


Fig. 4. Pycnidial conidioma of *Septoria hodgesii*; scale bar = 17 μm.

Other than *S. myricae* and *S. hodgesii*, the only species of *Septoria* described on a *Myrica* host appears to be *S. myricata* Dearn. & House (Dearness and House, 1940) on leaves of *M. gale* in New York State. *Septoria myricata* was described with "pycnidia opening amphigenously but chiefly above." Measurements of the pycnidial conidiomata were not given. Conidia were described as "obtuse, straight or somewhat curved, closely septate, the septa 3–7 μm apart, 15–33 × 3 μm." Dearness and House (1940) compared the description of *S. myricata* with that of *S. myricae*, emphasizing



Figs. 5–7. Conidia of *Septoria* spp. from *Myrica* hosts. 5. *S. hodgesii*; scale bar = 12 µm. 6. *S. myricae* from the type specimen of *M. cerifera*. 7. *S. myricata* from the type specimen of *M. gale*; scale bar for Figs. 6 and 7 = 10 µm.

the difference in spore size and stating that *S. myricae* was "otherwise different." My examination of material from original collections revealed that the leaf spots associated with infection by *S. myricata* differed visibly from those of both *S. myricae* and *S. hodgesii*. Leaf spots on *M. gale* appeared as large indistinctly delimited darkened blotches, with no necrotic tissue. The blotches, visible on both surfaces, occupied large portions of the leaf tissue (Fig. 3). As with *S. myricae,* few fertile conidiomata of *S. myricata* could be found, with only a meager number of conidia available for examination. Those seen, however, agreed morphologically with those described for this species, measuring 11–31 × 2–3 µm (Fig. 7). Septa were obscure, but those seen were 4–7 µm apart. Aside from the original material examined as described above, I am not aware of any additional collections of *S. myricae* or *S. myricata*.

Comparison of conidial, as well as other characteristics, of *S. hodgesii* with those of *S. myricae* and *S. myricata* clearly indicates the latter to be distinct species both from *S. hodgesii* and from each other. Although some of the bacillary conidia of *S. myricae* measured as long as 12 µm, these conidia are unusually small for a species of *Septoria* according to current concepts of the genus. Furthermore, septa were not visible in my examination, leading to questions as to the proper placement of this fungus.

The extent of distribution of *S. hodgesii* has not been determined. However, in Florida it is described as one of several frequently encountered leaf spot pathogens of wax myrtle both in natural stands and in cultivation (Schubert, 1986). C. S. Hodges (unpublished) has observed it commonly on wax myrtle in the coastal plains of North and South Carolina, as well as in commercial nurseries and ornamental plantings in North Carolina. It is likely that under environmental conditions conducive for disease development, leaf spots associated with this pathogen may be found throughout the range of *M. cerifera*.

Acknowledgements

I thank Amy Rossman and Rebecca Bennett of the U. S. National Fungus Collections, Scott Redhead of the National Mycological Herbarium of Canada, and John Haines of the New York State Museum for their helpfulness in supplying herbarium material. I am also grateful to Eloise Killgore and Lionel Sugiyama of the Hawai'i Department of Agriculture, and Rob Anderson of the Department of Botany, University of Hawai'i, for their technical assistance and support. I am grateful for the use of facilities and greenhouse space of the Department of Plant Pathology, North Carolina State University. I appreciate the helpfulness of Larry Grand in reviewing the manuscript.

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MYCOTAXON

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MICROPORELLUS OBOVATUS IN INDIA AND ITS CULTURAL CHARACTERISTICS

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ABSTRACT

Microporellus obovatus (Jungh.) Ryv., a poroid member of Aphyllophorales, is reported as being new to India. Its cultural characters are described for the first time. Detailed morphological and cultural characters of *Microporellus obovatus* are given based on the Indian collection and in the light of these observations the taxonomy of the fungus is discussed.

Key words : *Microporellus obovatus* (Jungh.) Ryv., Cultural studies, taxonomy, sexuality, India.

INTRODUCTION

During a mycological excursion in June 1996, several basidiocarps of a poroid fungus of Aphyllophorales were collected by the author. They grew on a dead stump of an unknown angiosperm at Darjeeling, West Bengal, India and were associated with a white rot. It was identified as *Microporellus obovatus* (Jungh.) Ryv. Perusal of literature (Bakshi, 1971; Bilgrami *et al.*, 1991) revealed that this report constitutes the first record of *Microporellus obovatus* from India. *M. obovatus* has also been found to grow in French Guiana, Guadeloupe (David and Rajchenberg 1985), Kenya, Nigeria, Sierra Leone, Tanzania, Zaire (Ryvarden and Johansen 1980) and USA (Gilbertson and Ryvarden 1987). Morphological and anatomical characters of this fungus were described earlier by some workers (Ryvarden and Johansen 1980, Gilbertson and Ryvarden 1987) but its cultural characters have not been studied before. In the present paper detail morphological and

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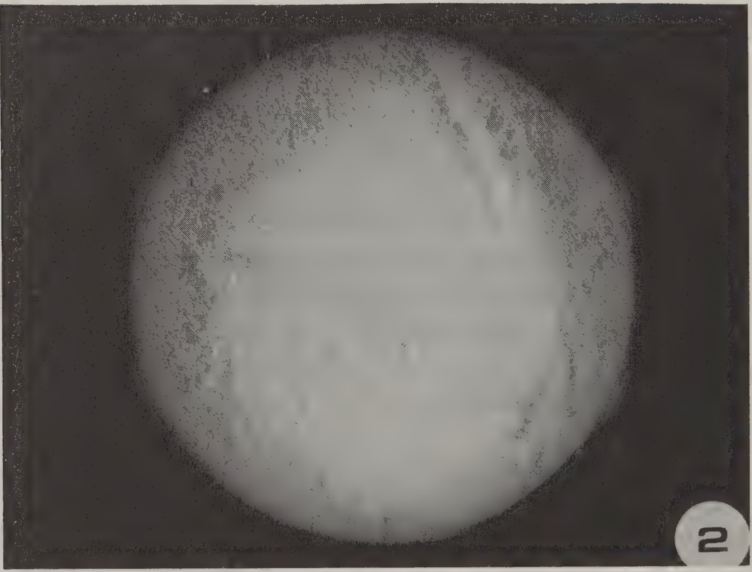

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cultural characters of *M. obovatus* are given based on the Indian collection.

MATERIALS AND METHODS

Observations were based on fresh specimens. Microscopic characters of the basidiocarps were studied from free hand sections mounted in 10% KOH and stained with 1% cotton blue. Cultures were established from the context tissues of these basidiocarps as well as from basidiospores. Cultural studies were made following the methods of Nobles (1948,1965) and Nobles *et al.* (1957). Oxidase reactions were tested by the Bavendamm method as described by Davidson *et al.* (1938). Amyloid reactions of basidiospores and dextrinoid reactions were determined in Melzer's reagent (Melzer 1924). Voucher specimens and culture have been deposited in the Mycological Herbarium, Department of Botany, Visva-Bharati University (VBMH), Santiniketan, West Bengal, India, a part of the specimens in the Herbarium of the Division of Mycology and Plant Pathology, Forest Research Institute (FRI), Dehra Dun, India and in the Mycological Herbarium, University of Oslo, Norway.

DESCRIPTION OF BASIDIOCARP

Basidiocarps annual, caespitose, rarely solitary, laterally stipitate with a small tapering stipe, pileus circular, reniform, spathulate or flabelliform, 2-5x2-3.2x0.2-0.5 cm. Pileus surface at first finely tomentose, becoming glabrous with age, radially folded with faint concentric zonations, white to pale buff in colour. Pore surface (Fig. 1) concolourous with the pileus surface, folded, pores angular to sinuous to daedaloid, 4-5 per mm, pore tubes up to 2.5 mm long. Context white, thin, up to 2.5 mm thick. Margin wavy, sterile, up to 2.5 mm broad.

Hyphal system dimitic. Generative hyphae hyaline, clamped, thin-walled (Fig. 3) to thick-walled (Fig. 4) to subsolid, 1.5-5.0 μm in diameter. The broken part of the thick-walled and subsolid generative

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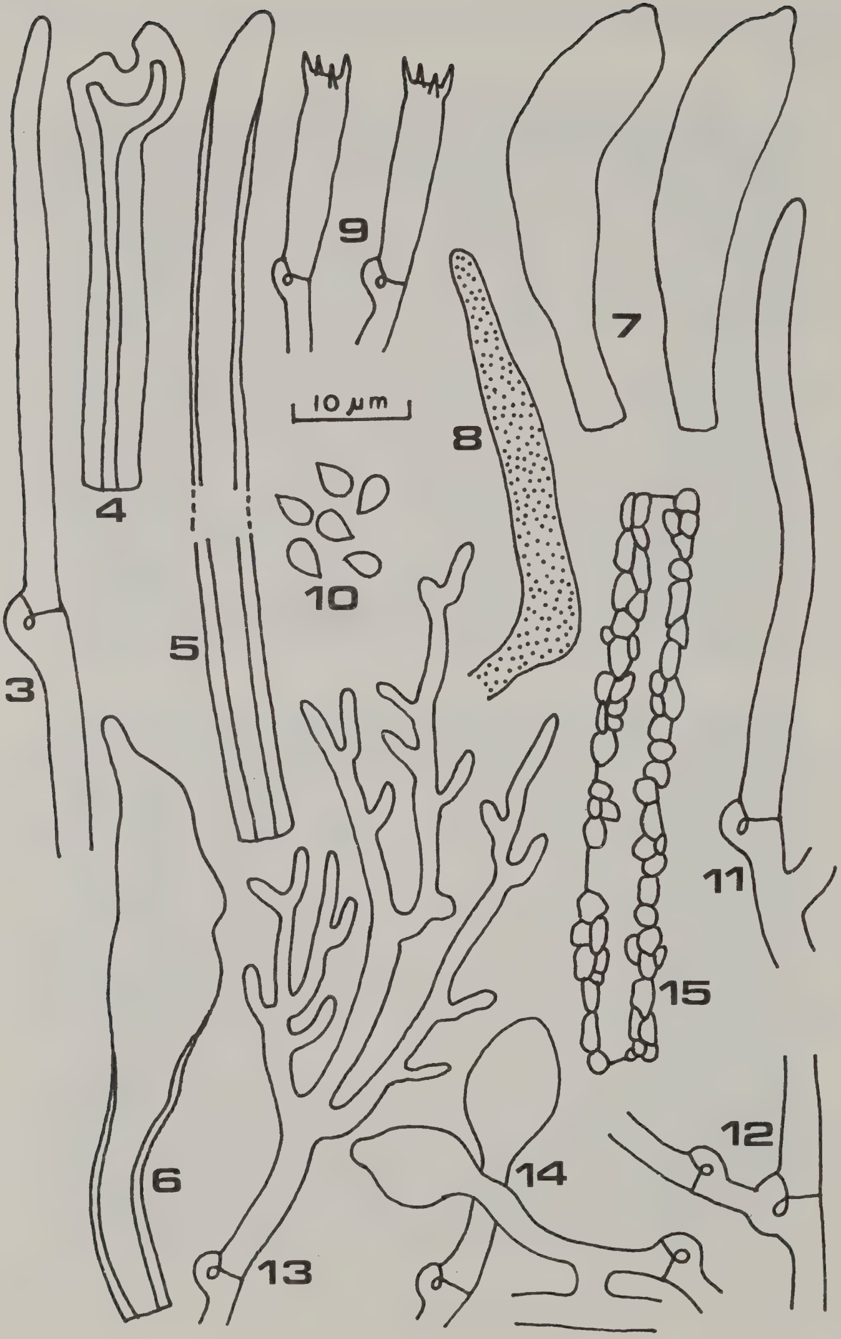
Figs. 1&2. *Microporellus obovatus* (Jungh.) Ryv. :1. hymenial surface of basidiocarp; 2. 6 weeks old mycelial mat in culture.


10 µm

hyphae may be mistaken for skeletal hyphae. Skeletal hyphae slightly to highly thick-walled, unbranched, 3-5 μm in diameter, dextrinoid (Fig. 5). Inside trama the tips of the skeletal hyphae bulge repeatedly (Fig. 6). This type of hyphal tips are also present in the hymenial layer forming cystidia (Fig. 7), up to 8 μm in diameter. Gloeocystidia (Fig. 8) are also present in the hymenial layer, thin-walled, up to 5.2 μm in diameter. Basidia (Fig. 9) clavate, 4-sterigmate, 14.5-20.3x2.8-4.2 μm. Basidiospores (Fig. 10) hyaline, thin-walled, globose to ellipsoid, smooth, 2.5-4.2 x 1.6-2.4 μm, non-amyloid.

ROT : Associated with white rot.

SPECIMEN EXAMINED : VBMH AMD 961, Darjeeling, West Bengal, India, 6500 ft. above sea level, on dead stump of an unknown angiosperm, 6 June, 1996.

DESCRIPTION OF CULTURE

Growth characteristics : Growth rapid, 4.5-5.0 cm in radius in the first week, covering the plate in two weeks. Advancing zone even, raised, white cottony mycelium extending to the limit of growth. Mat at first white, cottony to cottony-woolly, becoming light buff in the older region. Mat becoming woolly to woolly-felty with age (Fig. 2). Tuft of raised mycelium is present in the older region and mat becoming lacunose in some areas. In the older culture mycelium raised to the edge of the upper petri dish. Reverse bleached.

Oxidase reactions positive on both gallic acid and tannic acid agar. Colony 6.4-7.2 cm in diameter on both the media in 7 days. Diffusion zone moderately strong on gallic acid medium but faint on tannic acid medium.

Figs. 3-15. *Microporellus obovatus* (Jungh.) Ryv. 3-10. Microscopic structures from basidiocarp. 3. Thin-walled generative hypha. 4. Thick-walled generative hypha. 5. Skeletal hypha. 6. Swollen tip of skeletal hypha. 7. Cystidia. 8. Gloeocystidium. 9. Basidia. 10. Basidiospores. 11-15. Microscopic structures from culture. 11. Hypha from advancing zone. 12. Thin-walled aerial hypha with medalion clamp. 13. Aerial hypha with staghorn branches. 14. Thin-walled aerial hyphae with apical swellings. 15. Thin-walled encrusted hypha.

Microscopic characteristics : Advancing zone- hyphae hyaline, thin-walled, clamped, branched, 2.2-4.5 μm in diameter (Fig. 11).
Aerial mycelium — (i) hyphae hyaline, thin-walled, with medalion clamps, 2-3.5 μm in diameter (Fig. 12); (ii) hyphae hyaline, thin-walled, clamped, with staghorn branches, 2-3 μm in diameter (Fig. 13); (iii) hyphae hyaline, thin-walled, well branched, clamped, with H-connections and apical swellings (Fig. 14); (iv) hyphae hyaline, thin-walled, wide, 4-6.5 μm in diameter, encrusted (Fig. 15).

SEXUALITY : Result of interfertility tests of 20 monokaryotic cultures obtained from a fresh basidiocarp (VBMH AMD 961) show the species to be heterothallic and tetrapolar. The distribution of mating types among the basidiospores studied was as follows :

A_1B_1 : 1, 7, 12, 13, 14, 18
A_2B_2 : 3, 5, 6, 17
A_1B_2 : 4, 10, 11, 16
A_2B_1 : 2, 8, 9, 15, 19, 20

SPECIES CODE : 2.3.7.26.32.36.40.42.53.54.60 (following the system of Nobles 1965).

CULTURE EXAMINED : VBMH AMD 961.

DISCUSSION

According to David and Rajchenberg (1985) macro and micromorphological features of *M. obovatus* do not match with the definition of the genus *Microporellus* Murr. and this species should be excluded from the genus *Microporellus*. They also opined that its place among other genera of Polyporaceae is also uncertain and creation of a new genus should be preceeded by a careful study of other species included by Murrill in *Microporellus*.

But my study of basidiocarps and a culture revealed that *M. obovatus* resembles closely *Microporellus dealbatus* (Berk. & Curt.) Murr., the type species of *Microporellus* Murr. Like *M. dealbatus* (Gilbertson and Ryvarden 1987) *M. obovatus* causes white rot; possesses annual, stipitate basidiocarp with hairy pileus surface and poroid hymenial surface; bears dimitic hyphal system with clamped generative hyphae and unbranched dextrinoid skeletal hyphae, clavate, tetrasterigmatic basidia, ellipsoid, non-amyloid, smooth, hyaline basidiospores and cystidia. Besides, like *M. dealbatus* (David and Rajchenberg 1985) *M. obovatus* shows rapid rate of growth in culture, positive result in oxidase test and tetrapolar type of sexuality. Such a great number of similarities between these two species indicate a phylogenetic relationship.

Therefore, it is concluded that *M. dealbatus* and *M. obovatus* are congeneric and inclusion of *M. obovatus* in the genus *Microporellus* by Ryvarden (1972) is fully justified.

ACKNOWLEDGEMENT

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MYCOTAXON

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ALTERNARIA THEMES AND VARIATIONS (226-235)
CLASSIFICATION OF CITRUS PATHOGENS

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ABSTRACT

Alternaria isolates from leafspot of rough lemon (*Citrus jambhiri*) and brown spot of tangerine (*C. reticulata*) and tangelo (*C. paradisi* x *C. reticulata*) were studied from a morphotaxonomic viewpoint. Isolates originated in citrus-growing regions of Colombia, Israel, Turkey, South Africa, and the U.S.A. (Florida). Of the 135 isolates examined under controlled culture conditions, 77 are assigned to 10 new species of *Alternaria*. Not one of the 135 pathology-related strains can be identified morphologically with either typical *A. alternata* or typical *A. citri*. Notable here are "host-specific-toxin-producing pathotype" strains that have been misidentified for many years as "*Alternaria alternata.*"

The most abundant taxon from rough lemon (all Florida sources) is *A. limoniasperae* sp. nov. The most abundant from brown spot in Florida and Colombia is *A. tangelonis* sp. nov.; from brown spot in Israel, Turkey, and South Africa is *A. turkisafria* sp. nov. A second major group of isolates from rough lemon (Florida) is *A. citrimacularis* sp. nov. Other morphologically unique isolates described as spp. nov. are *A. citriarbusti* (tangelo, Florida), *A. toxicogenica* (tangerine, Florida), *A. colombiana* (tangelo, Colombia), *A. perangusta* (tangelo, Turkey), *A. interrupta* (tangelo, Israel), and *A. dumosa* (tangelo, Israel).

Keywords: *Alternaria*, citrus pathogens, host-specific toxins

XXVI. ALTERNARIA PATHOGENS OF CITRUS

Introduction

At least four well-known pathologies of cultivated *Citrus* species are incited by species of *Alternaria*. The fungus/disease relationships are widely recognized and probably occur wherever susceptible cultivars are grown. The *Alternaria* species associated with the diseases are less well known (with one exception, *A. limicola* Simmons & Palm (Simmons 1990)), primarily because isolates of the fungus incitants themselves have been characterized only superficially by concerned citrus pathologists.

No specialist in *Alternaria* taxonomy to date has made a concerted attempt to characterize large numbers of citrus-disease isolates from a wide range of geographic sources. And certainly no critical study relating morphologically distinguishable taxa of *Alternaria* to the recognized variety of associated citrus diseases has been published.

In consequence, most of the literature on strikingly different citrus diseases has been assigned uncritically to *Alternaria citri* Ellis & Pierce (in Pierce 1902), which originally was described only from an internal dry rot of orange fruit and whose type morphology has little in common with large numbers of isolates from other citrus diseases, as will be demonstrated below.

Additionally, and as a reflection of insufficient consideration of the microscopic characters of isolates, a few published proposals maintain that unique components of the citrus-related complex of small-spored *Alternaria* are morphologically indistinguishable from habitually misidentified *Alternaria alternata* (Fr. : Fr.) Keissler (1912). This position, which reduces the biological species concept to a matter of spore-size similarities or of statistical interpretation of DNA components, ignores the utility and validity of several names that carry with them considerably more and pertinent biological information than that relied on by the "*Alternaria alternata*"-covers-everything school of classification. That view is untenable for anyone who needs discriminative, differential information on the several different taxa. The view, unsupported by critical morphological data, is confronted in detail in a companion paper *Alt.T&V* 236-243 (Simmons 1999).

The four best-known *Alternaria* diseases of citrus are an internal dry rot of fruit, commonly called simply *Alternaria* rot of citrus, from a specimen of which *A. citri* originally was described; citrus leafspot (*mancha foliar de los citricos*), caused by *A. limicola*, described from Mexican (Key) lime (*C. aurantiifolia* (Christm.) Swingle), and reported from Mexico on other *Citrus* species as well (Palm and Civerolo 1994); *Alternaria* brown spot of tangerine (mandarin; C. *reticulata* Blanco) and related hybrids and cultivars; and

Alternaria leafspot of rough lemon (*C. jambhiri* Lush.).

Typical *Alternaria citri* is known from miscellaneous collections of citrus fruit, leaves, and twigs (see Simmons 1990, *Alt.T&V* no. 28). Judging from a few isolates derived from fruit dry rot that have been available to me, *A. citri* is not uniquely responsible for this widespread condition of developing and postharvest fruits. The dry-rot condition is particularly prevalent on navel oranges, but has been reported also from lemon, lime, tangerine, grapefruit, and their hybrids (Brown and Eckert 1988; Farr et al. 1989). It is not discussed further here.

Typical *A. limicola* causing leafspot of lime and other citrus species, the subject of studies by Simmons (1990 *Alt.T&V* 27) and by Palm and Civerolo (1994), also is not discussed further here.

The morphotaxonomy of *Alternaria* species that are linked with leafspot of rough lemon and with brown spot of tangerine and related cultivars is the focus of the remainder of these observations. After a survey of the taxonomic and nomenclatural history of associated *Alternaria*, they are treated separately below.

Historical Background

According to K. G. Pegg (1966), *Alternaria* brown spot of citrus was first described on Emperor mandarin in Australia in 1903 (Cobb). Pegg (1966) contributed an early study of the disease, the host-specific nature of the toxin(s) produced in filtrates from cultures of pathogenic *Alternaria* isolates, and comparisons of conidium dimensions of his isolates from Australian sources. The symptoms subsequently have been recognized in association with fruit and foliage pathologies of major importance in most citrus-growing areas of the world. The disease appeared suddenly and destructively in 1973 in Florida, U.S.A. (Whiteside 1976a and b), where it has become a major problem on tangerine and tangelo cultivars. Estimates of loss vary, but are large, with a report from Colombia for 1993, for example, stating that the disease affects 90% of citrus cultivars grown in several areas of that country (Castro et al., 1994.)

Alternaria isolates associated with *Alternaria* brown spot usually have been referred to as *Alternaria citri* Ellis & Pierce; or as *A. alternata* (Fr.) Keissler (= *A. tenuis* Nees in earlier literature); or, by individuals honestly acknowledging the limitation of their taxonomic expertise, simply as *Alternaria* sp. J. O. Whiteside (1988), the leading authority on the subject for many years, held that "the causal agents of [brown spot of mandarin and of leaf spot of rough lemon] ... differ from the fungus ... causing black rot" [i.e., *A. citri*]. Nevertheless, he succumbed to the lure of so-called "recent taxonomic criteria" and suggested that "the brown spot pathogen should probably be regarded as a distinct pathotype of the collective species *A. alternata* (Fr.) Keissler."

Whiteside's "recent taxonomic criteria" referred to an unsupportable practice, becoming fashionable at the time, of treating readily distinguishable small-spored taxa of *Alternaria* simply as a group of toxin-producing "pathotypes" within *A. alternata*. The reasoning was that "these fungi look similar in conidial morphology, and statistically the measurements of conidial size fall within a category of the descriptions of a collective species *Alternaria alternata* (Fries) Keissler, as described by Simmons [1967]" (Nishimura and Kohmoto 1983; paraphrasing Nishimura 1980; in turn based on Nishimura et al. 1978).

This viewpoint of the Nishimura school is indefensible in the 1980-90s era of cultures compared under specified, controlled conditions and of evolving molecular studies; it has been indefensible since its inception in 1978.

For the record, let it be emphasized that Simmons did not in 1967 describe, nor has he ever, at any time in the subsequent 30 years of his *Alternaria* studies, published, held, or voiced a "collective species" concept for the name and species *Alternaria alternata*. The Nishimura et al. papers cited in a preceding paragraph are patently incorrect in their statements; exhibit a lack of understanding of what Simmons actually presented in that 1967 paper (formal neotypification of the name *A. alternata*); and in the years 1978 to date have managed to confuse practicing mycotaxonomists and species-knowledgable pathologists into believing that there is some defined morphologic basis underlying the use of a "collective species" concept of typical *Alternaria alternata*. There is not. The subject is pursued in detail, with specific examples, in a companion paper *Alt.T&V* 236-243 (Simmons 1999).

The second disease discussed here, *Alternaria* leaf spot of rough lemon, was first described in South Africa in 1929 (Doidge). The disease is present in many areas where rough lemon and Rangpur lime (*C. limonia* Osbeck) are grown or used as rootstock for other citrus species (Whiteside 1988). An early report of the disease for Florida (Ruehle 1937) considered the instigating *Alternaria* to be morphologically similar to fruit-rotting *A. citri*, but cited data showing that isolates from rough lemon and ones from dry rot did not produce comparable symptoms in cross-inoculation work with the two hosts. Doidge (1929) had published similar observations, noting that only *Alternaria* strains from rough lemon attacked rough lemon seedlings, and that none of the isolates she used from orange induced lesions in rough lemon leaves. Similar results and conclusions, based on toxic filtrates applied to tangerine and rough lemon leaves, were reached by Kohmoto, Scheffer and Whiteside (1979).

In summary, pathologists—from the earliest recognition of the brown spot and rough lemon pathologies up to the present time—have reported that at least two demonstrably distinct *Alternaria* taxa are operative in two quite different citrus problems. For the most part they have used the name *A. citri*, incorrectly, as an umbrella for the specimens and isolates they have handled.

Current Project

During the July 1996 joint meeting of the American Phytopathology Society and the Mycological Society of America, Dr. Mary E. Palm (Animal and Plant Health Inspection Service, USDA, Beltsville, Maryland) introduced citrus pathologist L. W. Timmer to E. G. Simmons; the three discussed the potential benefit of adding a dimension of taxonomic opinion to Timmer's ongoing citrus pathology program at the University of Florida's Citrus Research and Education Center, Lake Alfred, Florida.

Optimism was in order, as Timmer and T. L. Peever, his colleague at the time at Lake Alfred, had underway a project primarily on epidemiological aspects of citrus diseases and related questions of pathogen specificity in different growing regions worldwide.

Additionally, Simmons fairly recently had published on the identity and taxonomic status of the 20 names of dark, dictyospored hyphomycetes described from *Citrus* and other Rutaceae (1990; *Alt.T&V* 27-53). At that time he had avoided the taxonomic bucket-of-worms comprising identifications of relatively small-spored *Alternaria* species associated with major citrus diseases, for three reasons:

1. the body of pertinent literature contained little definitive information on the morphology of the *Alternaria* taxa involved in the diseases;

2. Simmons did not at the time have in hand a

representatively large number of well-documented *Alternaria* isolates from citrus hosts;

3. the name *Alternaria citri* had been adopted indiscriminately over the years for *Alternaria* isolates related to quite dissimilar, even cross-inoculation-negative citrus pathologies. And even more confusing to pathologists and mycotaxonomists, the names *Alternaria alternata* and *A. citri* were being promoted as catchalls for several widely recognized and destructive *Alternaria* pathogens, solely on the basis of remarkably unsophisticated morphological criteria (Bottalico and Logrieco 1992; Nishimura et al. 1978; Nishimura 1980; Nishimura and Kohmoto 1983; Otani and Kohmoto 1992; Scheffer 1992; Scheffer and Livingston 1984).

A plan was devised to channel contributions of pathogenic *Alternaria* isolates to Simmons from cooperating citrus pathologists in several different countries. These would be added to ex-citrus isolates already in his hands from previous donors. No information on specific disease or involved tissue would be given to Simmons until initial morphotaxonomic comparisons had been completed by him. The only assumption available to him was that each isolate had already been shown to be pathogenic to some citrus host. Inoculation and molecular studies of isolates at the Lake Alfred facility were not to be shared with him until after his initial opinions on sporulation and microscopic groupings had been established.

Isolates Assembled for Morphotaxonomic Study

Florida (1990)	9 Whiteside isolates dating from 1974 (his 300-series) and 1990 (his 700-series), from tangerine & rough lemon
Colombia (1994)	19 isol., all from Minneola tangelo (*C. reticulata* x *C. paradisi* Macf.)
Colombia (1996)	7 isol., all from Minneola tangelo
Israel (1996)	13 isol., all from Minneola tangelo
South Africa (1996)	7 isol., all from Minneola tangelo
Turkey (1996)	10 isol., all from Minneola tangelo
Florida (1996)	17 isol. from tangerine (*C. reticulata*), tangelo, tangor (*C. sinensis* x *C. reticulata*), rough lemon (*C. jambhiri*)

Florida (1997)	31 isol. from rough lemon
Florida (1998)	15 isol. from tangelo
Florida (1998)	1 additional Whiteside isolate (PR309) from rough lemon.

It is worth noting at this point, as certainly is done elsewhere (below, and Simmons 1999), that some of the Whiteside strains, isolated in the 1970s, have been the basis of work on host-specific toxins and phylogenetic analysis of *Alternaria* isolates from tangerine and rough lemon, and thus indirectly of the unjustifiable belief that these "citrus pathotypes" are morphologically indistinguishable from *A. alternata* (Kohmoto et al. 1991; Kusabe and Tsuge 1995).

Because typical *Alternaria alternata* and *A. citri* are mentioned frequently in these discussions, illustrations of their sporulation patterns and representative conidia are inserted for reference on the next page (Fig. 154).

Protocols, Caveats, Recommendations

The working collection of isolates comprised two major groups: 44 from rough lemon, and 91 from Minneola tangelo and a few other tangerine-related cultivars.

All cultures underwent reisolation by Simmons by means of 1-conidium or 1-chain selections. All isolates were studied following growth in 90mm Y-plates under identical conditions of agar substrate, temperature, humidity, and light exposure, viz., PCA, Hay, V-8 in sectors of each plate; 22-24C; ambient, gradually drying atmosphere; cool-white fluorescent light, cycled daily 8hr light/16hr dark (see Simmons 1992, Simmons and Roberts 1993 for details). Observations of sporulation patterns were made at 50X in plates and, at higher magnifications, in lactic acid on double-face sticky tape preparations from active cultures.

It has been my practice in earlier *Alt.T&V* notes to describe and compare species on the basis of potato-carrot agar (PCA) cultures. In a few instances comparisons were made from V-8 juice agar because this medium accentuated sporulation differences that were less obvious on PCA (e.g., for species similar to *A. tenuissima* in *Alt.T&V* 117-122 (Simmons 1995)).

The varied but distinctive morphologies exhibited among the present set of ex-citrus isolates led me to emphasize whichever of the three standard media supported the firmest

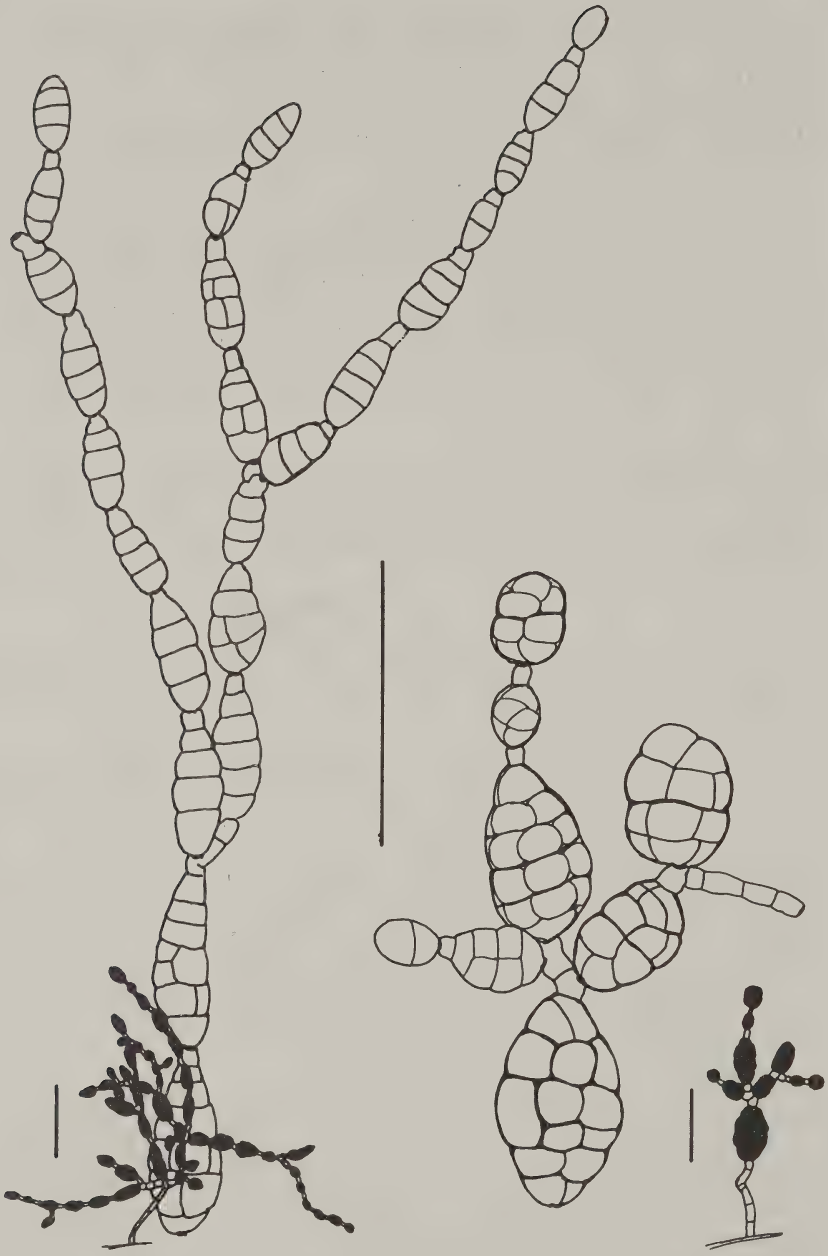

Fig. 154. Sporulation patterns and mature conidia in part of sporulation apparatus: left, *Alternaria alternata* ex representative EGS 34-016; right, *A. citri* ex type specimen EGS 07-106. Bars = 50μm.

diagnosis. In this instance descriptions and illustrations are provided of preparations from all three media so that comparisons can be visualized more readily. The type specimen for each species was prepared, however, from the single medium cited in its latinized type diagnosis.

Here again, as has been done many times previously, I recommend that these three media be used routinely by others attempting to study and characterize *Alternaria* isolates. A relatively simple standardization of this sort would help immeasurably in communication among students of the genus.

Descriptions in the text are based on cultures 5-6 days old; comments and illustrations based on younger or older colonies are identified as such. Since conidiophore development and sporulation usually are affected by the daily light/dark cycle, comments related to this phenomenon are indicated in the text by phrases "light exposed" or "light phase," and "dark phase."

Fresh conidia of isolates of small-spored *Alternaria* species usually exhibit a pale gray-green hue that masks underlying variants of pale tans, browns, and near blacks. Preparations in clear lactic acid (mounting medium of choice) gradually lose this gray-green cast, quickly or within a few days, and expose the characteristic colors that remain stable in sealed slides for many years. All descriptions given here are from color-stabilized preparations.

To avoid the confusion of citing code numbers from various strain-numbering systems of donors, my study numbers, in the format EGS 00-000, are cited throughout the text. An Appendix preceding Literature Cited lists these study numbers along with equivalent source and strain information supplied by original contributors of isolates.

Alternaria Isolates from Rough Lemon Lesions

Assigning rough lemon (RL) isolates to morphologically recognizable groups proved to be an early encouraging exercise. Of 28 RL isolates in an initial set received in 1997, 16 were readily identifiable as a single morphologic taxon, which was quite distinct from any of the numerous (91) tangerine-related cultures, and which, as indicated below, is identical with Whiteside strains with a history of pathogenicity. Examination of 9 additional recent RL isolates from Florida yielded three more of these distinctive RL strains, plus three which were morphologically close but culturally poor fits. Two of the five Whiteside RL isolates dating from the 1970s also matched the distinctive RL group, as did one of two 1996 RL isolates from Florida.

This definable group of 22 of the 44 RL isolates, all from Florida, represents a taxon that is readily distinguishable from all others of the 135 strains available to this study. Notably, the two early Whiteside isolates that fit this distinct RL group (his PR309 and PR325) are ones cited in host-specific toxin (HST) studies by Kohmoto et al. (1991, for PR325 = EGS 39-185) and in phylogeny of HST strains by Kusabe and Tsuge (1995, for PR309 = ATCC 38962 = EGS 46-159).

Within the context of small-spored *Alternaria*, and certainly among the so-called host-specific pathotypes, this species has no more than a generic resemblance to typical *Alternaria alternata*, or to typical *A. citri*, or to any other of the taxa purported to be morphologically indistinguishable by the HST school of classifiers (see references above in "Current Project," item 3.)

This group of 22 RL isolates is associated with inoculation pathogenicity (Whiteside 1978, 1988) and host-specific-toxin data (Kohmoto et al. 1991). Its members are morphologically distinct among small-spored *Alternaria* known from citrus substrates as well as from any other source known to Simmons. The taxon is presented here as the previously undescribed *Alternaria* leafspot pathogen of rough lemon. It traditionally has been misidentified as *Alternaria citri* (Whiteside 1988) or, with negligible morphologic consideration, as an *A. citri* rough lemon "pathotype," a word coinage that contributes little to information content in *Alternaria* systematics or to any practical need for identification of the fungus.

The other 12 isolates in the original set of 28 from RL lesions have no published history of pathogenicity to rough lemon. They are a morphological group distinct from RL-pathogenic *A. limoniasperae* (*Alt.T&V* 226, below) and from the numerous isolates from Minneola tangelo. This group of 12 isolates supports a second new species *A. citrimacularis* (*Alt.T&V* 227, below). The balance of 10 RL isolates are a morphological miscellany that is not treated further here.

226. **Alternaria limoniasperae** Simmons, sp. nov. Fig. 155

Etym.: L. (gen.), *limon-* + *asper*, of rough lemon

Ex cultura in agaro Hay descripta. Coloniae modice concentrice zonatae, abunde conidiogenae. Conidiophora primigena brevia. Catenae conidiorum longissimae et per-

*angustae, constate ex conidiis 10-12 et 10-30. Conidia incremento praecoci longiangusta, 25-40 x 7-11µm, conidiophoris secundariis nunc 2-4 x 2-3µm nunc typice ad 25-50 x 2-3µm, 3-7 transverse, raro 1 longitudinaliter septata, dilute luteobrunnea, laevia. Habitatio typi in laesione Citrus jambhiri Lush., in arbusto citreo, Florida, U.S.A. Typus: partes ex cultura EGS 45-100 (isol. HC2-RLR-11s ex CREC, Lake Alfred, Florida; accept. vii.1997) desiccatae et in **BPI** (holotypus) et saltem in **IMI** (isotypus) conservandae.*

Colonies show concentric rings of growth and sporulation on all media used. Rings are only moderately apparent on PCA and Hay because conidiophores from both light-exposed and dark-phase areas are equally and abundantly conidiogenous. On V-8, strong zonation (light exposed) contains short conidiophores and abundant conidia and alternates with rings (dark phase) of very long, branching primary conidiophores that terminate in relatively short chains of conidia.

Primary conidiophores (light exposed) are short on all media, up to ca. 100µm, but usually shorter on Hay. In dark-phase zones, particularly on V-8, long suberect hyphae that act as primary conidiophores have several lateral branches and often 2-3 branches arising near the apex. All such branches are conidiogenous. The central suberect axis may be up to 0.5-1.0mm x 5µm; the branches are 50µm or more long.

On **PCA, conidial chains** typically have a primary axis of 4-6 units (at 2d), 10-12 units (5d), up to ca. 20 units (7-10d), and very commonly 1-4 lateral branches 4-10 spores long.

On **PCA, conidia** are narrow-ellipsoid (30-50 x 8-10µm) or ovoid (20-35 x 8-12µm) in the initial lower part of the chain but become ovoid and considerably smaller (8-12 x 4-8µm) in the terminal part of the chain. Conidia have very small secondary conidiophores (ca. 2-4 x 2-3µm) that often are reduced morphologically to only a conidiogenous pore in the otherwise undifferentiated apex of the preceding conidium; have 1-4 transepta and 1(-2) longisepta in relatively few of the conidia in any chain; are medium gold-brown and punctulate to punctate.

On **Hay**, which is used in typification of the species, **conidial chains** are very long and narrow, with 10-12 units (at 4d) and ca. 10-30 units as the colony ages. Chains usually are solitary, but sometimes a short primary conidiophore branches 2-3 times and gives rise to a tuft of 3-4 long chains. Early sporulation (5-6d) is diagnostic, in that

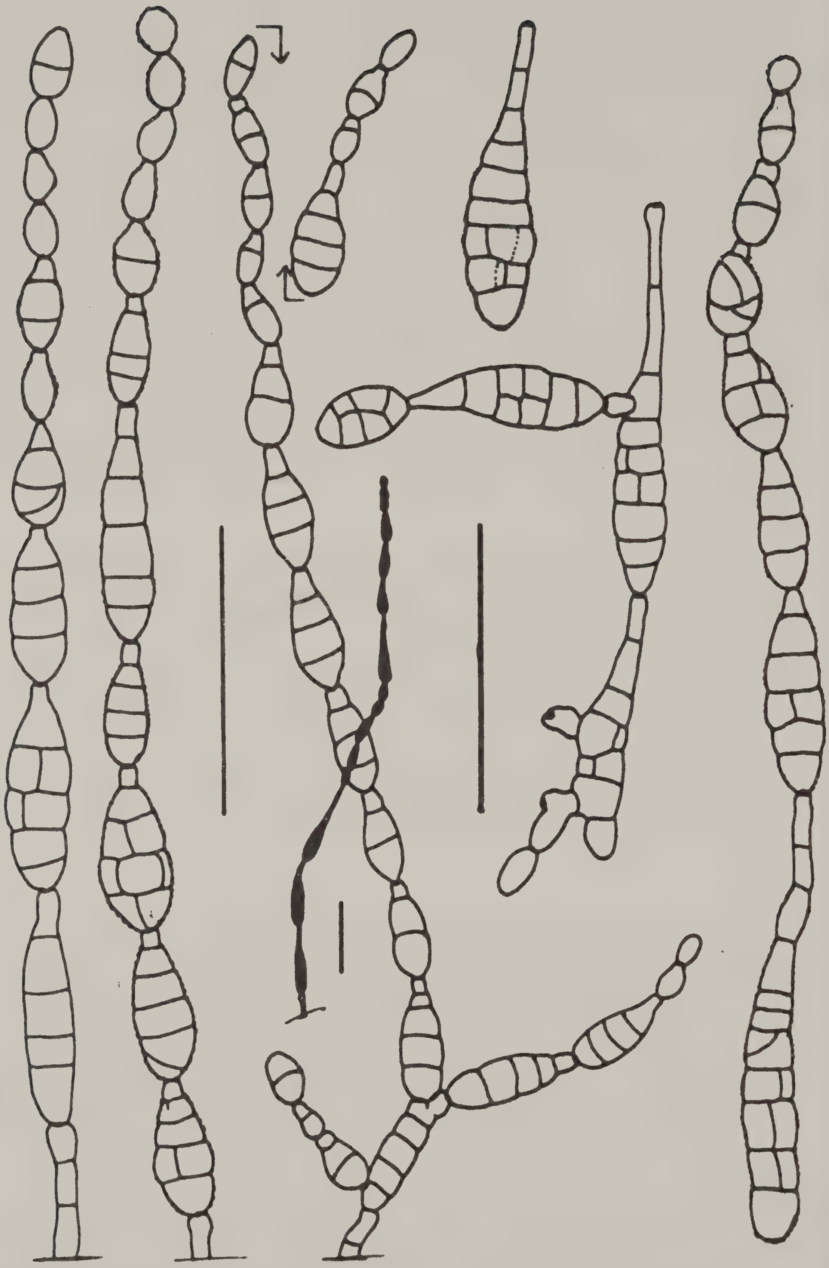

Fig. 155. *Alternaria limoniasperae*: conidia, conidiophores, habit ex type isolate EGS 45-100. At left, 6d. on PCA; at center, 10d. on Hay; at right, 6d. on V-8. Bars = 50μm.

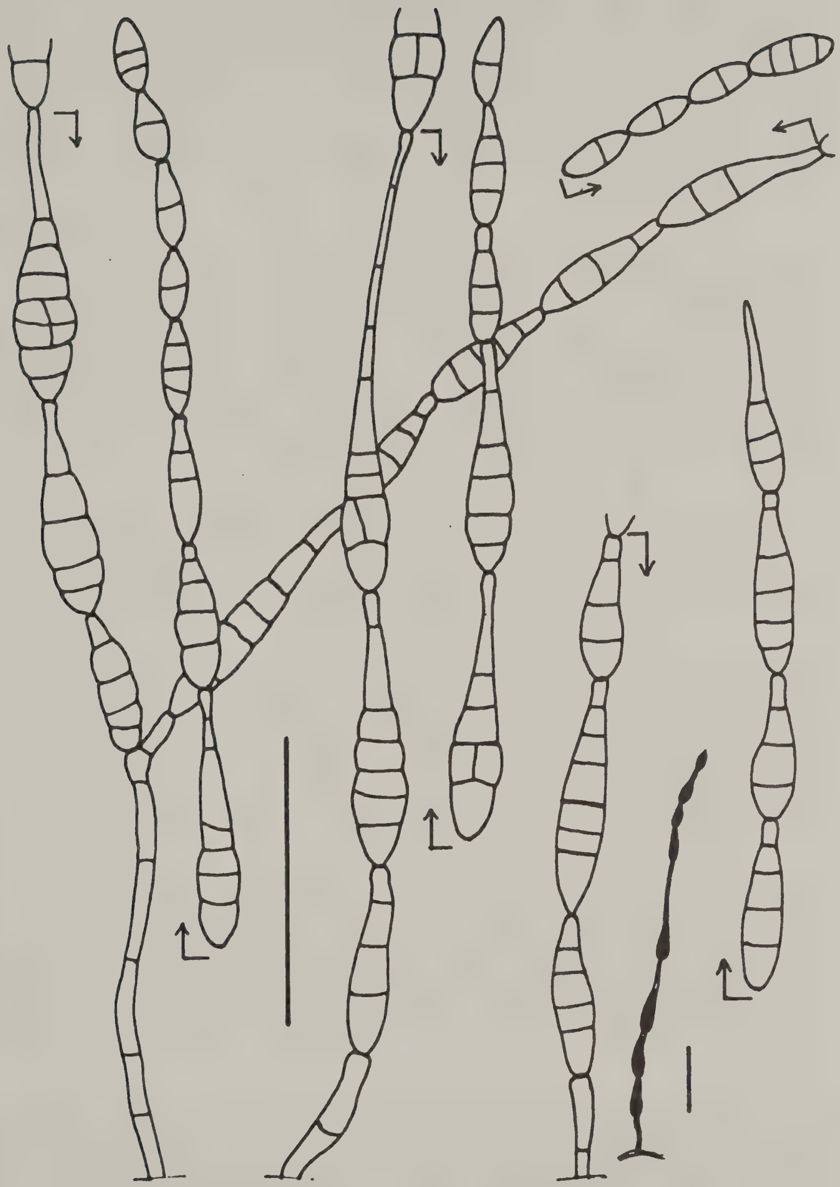

Fig. 155 contd. *Alternaria limoniasperae*: conidia, conidiophores, habit ex type isolate EGS 45-100; 6d. on Hay. Bars = 50μm.

chains appear narrow and fragile due to the narrow width of the conidia, the absence of longitudinal septa from most conidia, and occasionally the inclusion of one or two conidia with a narrow apical extension 1-2 times as long as its spore body. Chains produced later (7-10d) still may include a conidium with a somewhat long secondary conidiophore, but are composed increasingly of short, small conidia (8-15 x 5-8μm) in numbers approaching the maximum of ca. 30 per narrow, delicate chain.

On **Hay, conidia** in early growth are ca. 25-40 x 7-11μm; have secondary conidiophores mostly ca. 2-4 x 2-3μm but in some conidia up to 25-50 x 2-3μm; and 3-7 transepta plus a single longiseptum in 1-2 cells of only 1-2 conidia of a long chain. In later growth the short, small conidia have secondary conidiophores up to 7-25 x 2-3μm in oldest spores but only ca. 3 x 2-3μm in great numbers of the population. Most of the small conidia have 1-3 transepta and no longisepta. Conidia are a dilute yellow-tan and smooth.

On **V-8, conidial chains** (light exposed) of ca. 10-15 units are borne in crowded, entangled stands. Chains on longer branching conidiophores (dark phase) consist of 5-10 conidia produced at each conidiogenous site. Chains on V-8 have the same general pattern of initial relatively large conidia progressing into a terminal region of tiny spores that is characteristic of the longer chains produced on PCA and Hay. On **V-8, conidia** are narrow-ellipsoid up to ca. 30-55 x 11-12μm, to narrow-ovoid or ovoid to 10-30 x 5-10μm; have secondary conidiophores 2-25 x 2-3μm; have 1-8 transepta and 0-1 longisepta in 1-3 of a few conidia in any chain; and are medium gold-brown and punctulate to punctate overall.

All 20+ of the isolates identified as *A. limoniasperae* were isolated from leafspots of rough lemon. They have the same substrate and locale origins as a group of isolates discussed below (*Alt.T&V* 227) as *A. citrimacularis*, but the two species are readily differentiated from each other on any one of the standard media. See *Alt.T&V* 227 for a discussion of morphological differences emphasized by growth on Hay agar.

The typical and diagnostic very long narrow chains of *A. limoniasperae* conidia on Hay agar are approached in appearance within this set of 135 isolates only by those of a different taxon isolated from Minneola tangelo in Turkey. This latter species, with even finer, more delicate chains of very narrow conidia, is introduced below as *Alternaria perangusta* (*Alt.T&V* 233).

A list of *A. limoniasperae* isolates examined includes EGS 39-185/-187, 45-080/-081/-082/-083/-086/-088/-092/-094/-095/-097/-098/-099/-100/-101/-105/-107, 46-033/-036, and 46-159. See Appendix for original source information.

227. **Alternaria citrimacularis** Simmons, sp. nov. Fig. 156

Etym.: L., *citrus* + *macula* + *-aris*, related to citrus lesions.

Ex cultura in agaro V-8 descripta. Coloniae fortiter concentrice zonatae, dense conidiogenae. Conidiophora primigena brevia, 100-150 x 4-5µm (in luce), vulgo 5-10X longiora (in statu obscuro). Catenae conidiorum (in luce) constatae ex conidiis 8-15, ramulis brevibus. Conidia anguste ovoidea vel anguste ellipsoidea, 30-50 x 7-12µm, conidiophoris secundariis 3-5 x 4µm vel ad 16-35 x 3; 5-7 transverse et 1-4 longitudinaliter septata, modice aureo-brunnea, laevia vel punctulata. Habitatio typi in laesione Citrus jambhiri Lush., in arbusto citreo, Florida, U.S.A. Typus: partes ex cultura EGS 45-090 (isol. BC2-RLR-32s ex CREC, Lake Alfred, Florida; accept. vii.1997) desiccatae et in BPI (holotypus) et saltem in IMI (isotypus) conservandae.

Colonies exhibit concentric rings of growth and sporulation on all media used. Rings are only moderately apparent on PCA and Hay as conidiophores from both light-exposed and dark phase areas are equally and abundantly conidiogenous. On V-8, dense black rings (light exposed) of relatively short conidiophores and masses of conidia alternate with rings (dark phase) of very long, branching primary conidiophores that terminate in relatively short chains of conidia.

Primary conidiophores are relatively short on all media in light-exposed zones, up to ca. 100-150 x 4-5µm. In dark-phase zones, the structures are commonly 5-10 times as long; they are abundant but not crowded on PCA and Hay, and form an open but discrete stand of erect, branching conidiogenous elements on V-8.

On **PCA**, **conidial chains** rarely remain simple. They typically have a primary chain of 8-15 elements with lateral branches of 1-6 units. Branching and subsequent chain proliferation originate as lateral secondary conidiophores from conidium body cells or as geniculate extensions of primary conidiophores past terminal conidiogenous sites.

On **PCA, conidia** typically are narrow-ellipsoid, 30-40 x 7-10µm, with secondary conidiophores 3-8 x 2-3µm or occa-

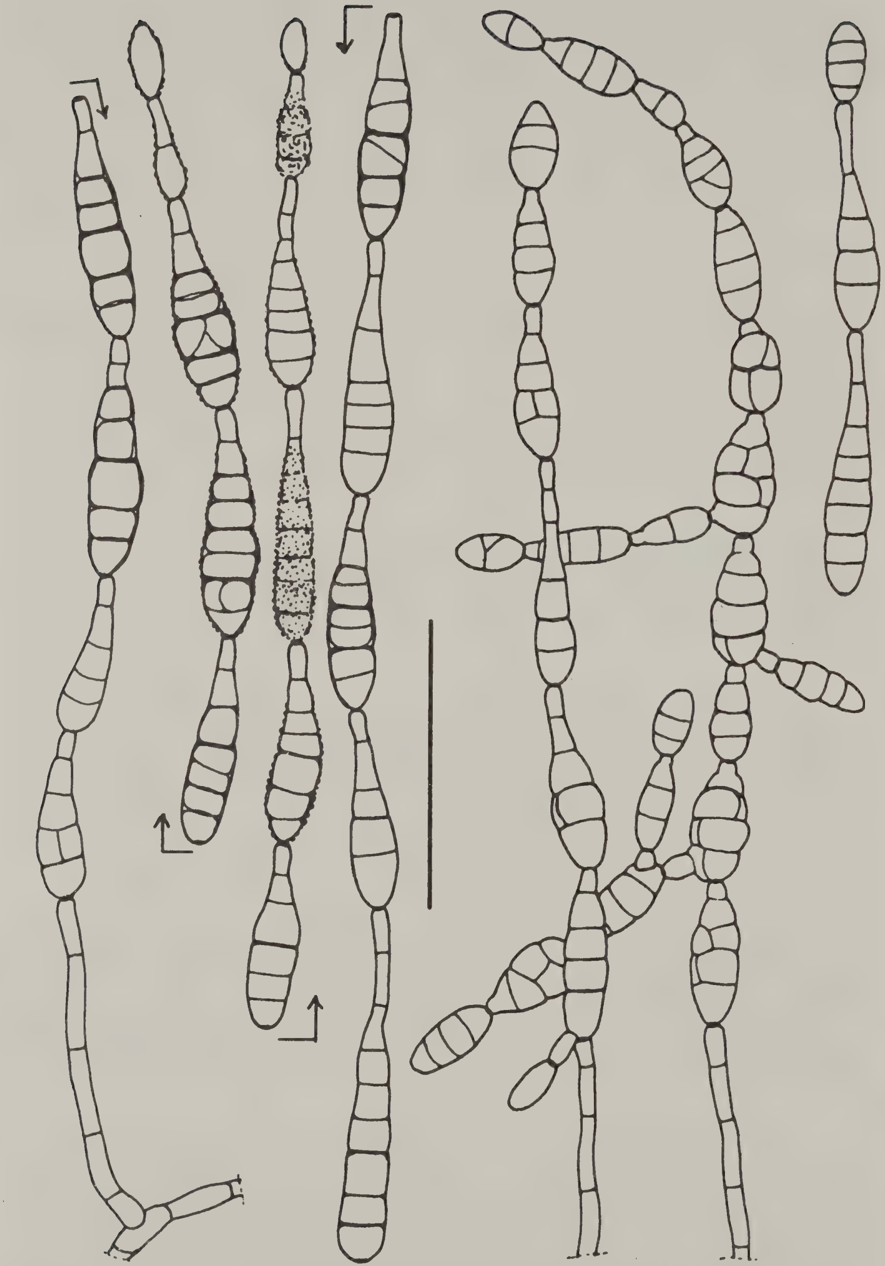

Fig. 156. *Alternaria citrimacularis*: conidia and conidiophores ex type isolate EGS 45-090. At left, 6d. on PCA; at right, 6d. on Hay. Bar = 50μm.

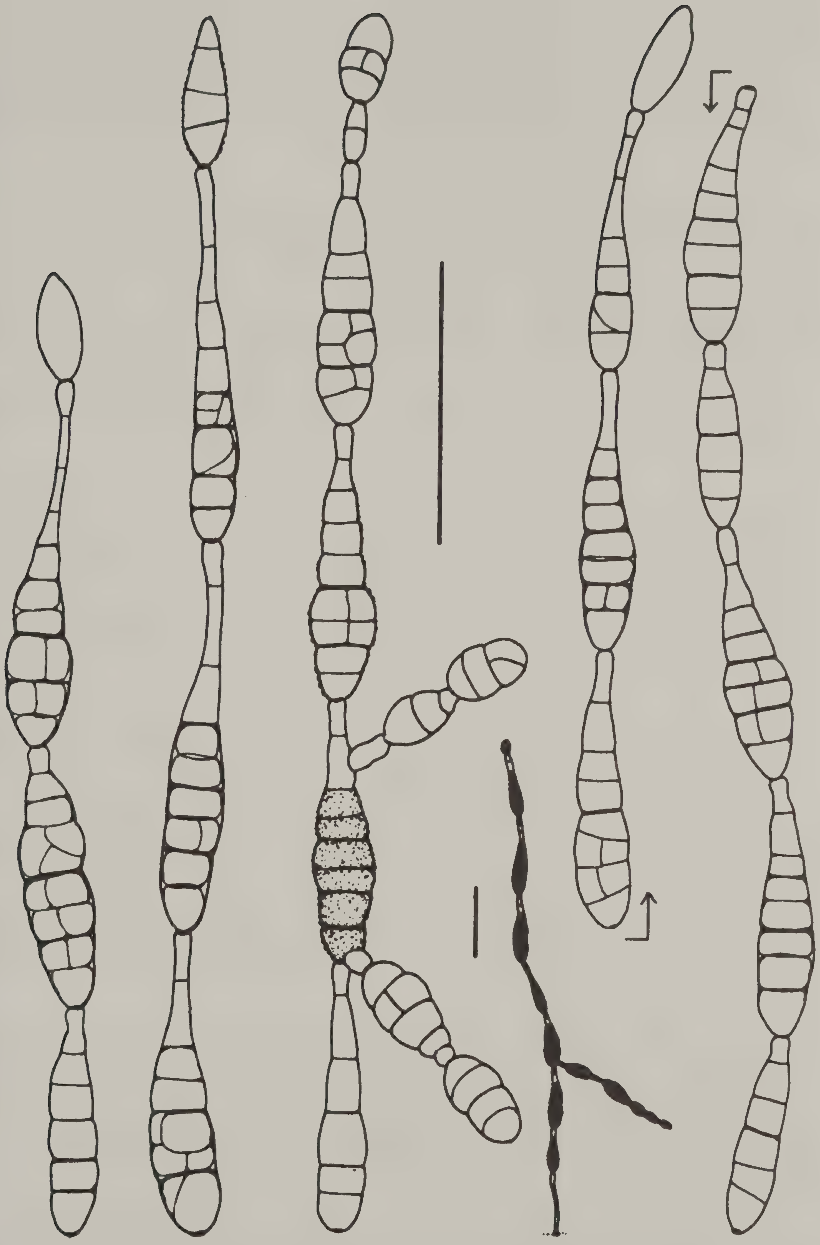

Fig. 156 contd. *Alternaria citrimacularis*: conidia, conidiophores, habit ex type isolate EGS 45-090; 5-7d. on V-8. Bars = 50μm.

sionally up to 20-25μm long in young growth; have 5-7 transepta and no longisepta, or at most 1 longiseptum in 1-3 of the transverse segments of a few conidia. As the colony ages to 1 wk, considerable numbers of short-ovoid conidia ca. 26-39 x 14-18μm are produced. Conidia are medium golden-brown and smooth to punctulate overall.

On **Hay, chains** are shorter than on PCA. They commonly are a simple 8-10 units long but more frequently develop as small clusters composed of the primary chain and 1-4 short lateral chains of 2-4 conidia each. Conidia exhibit narrow-ellipsoid and broader ovoid shapes, even in the same chain; secondary conidiophores usually are ca. 2-5 x 3μm, but occasionally are long-narrow, 25-35 x 2.5-3.0μm; have 1-6 transepta and 1 or no longisepta in narrow conidia, or 1 longiseptum in 1-3 of the transverse segments of ovoid conidia; are dilute yellow-tan and usually smooth.

On **V-8**, which is used in typification of the species, **chains** produced in light-exposed zones are similar to those on PCA and Hay, i.e., short to moderately long, with lateral branches; sporulation elements are so densely crowded that they form a black thicket of conidial chains. Chains on the much taller primary conidiophores and branches of the dark-phase zones are short, ca. 5-6 conidia, with lateral branches only 1-2 conidia long.

On **V-8, conidia** are narrow-ovoid to narrow-ellipsoid, 30-50 x 7-12μm; have secondary conidiophores 3-5 x 4μm, but also frequently ca. 16-35 x 3μm; have 5-7 transepta (the median euseptum often slightly darker and more constricting than others), and 1 longiseptum in 1-4 of the transverse segments; are medium gold-brown and smooth to punctulate overall. Increasing numbers of ovoid conidia ca. 30 x 15μm and narrow-ellipsoid conidia no longer than 40μm are produced as the colony ages near 7d, but the population picture remains predominantly that of long-narrow-ellipsoid conidia.

All 11+ isolates of my working set that represent *A. citrimacularis* are from leafspots of rough lemon. Although they have the same substrate and locale origins as other isolates from rough lemon that are treated here as *A. limoniasperae* (*Alt.T&V* 226), comparison on any of the standard media emphasizes their morphologic differences.

Hay agar is especially useful for differentiating the two species, even at the 50X level. On Hay, conidial chains of *A. limoniasperae* are long (10+ conidia), narrow-fragile in appearance, and typically include individual conidia with very long apical extensions 2-4 times longer than the spore body. In contrast, conidial chains of *A. citrimacularis* may have as many as 10 spores in chains but the conidia are sufficiently short and broad-ovoid, and produce enough lateral chains, that the appearance at 50X is more robust, with interconidial connective cells never strikingly long-narrow.

Pursuing comparison at higher magnifications reveals that the 50X differences are supported by conidium morphology: dominantly long-narrow in *A. limoniasperae*; shorter, dominantly broad-ovoid in *A. citrimacularis*.

A list for the record includes isolates (all from leaf lesions of rough lemon): EGS 45-084/-085/-087/-089/-090/-091/-103/-104/-106, 46-077 and 46-109. See Appendix for original source information.

Alternaria Isolates from Tangerine-related Cultivars

Grouping the 80-90 usably stable isolates from *Citrus reticulatus* hybrids and cultivars (mostly from Minneola tangelo) into morphologically recognizable taxa was a tedious, sometimes discouraging exercise. It quickly became apparent that no system or weighting of opinions based on sporulation patterns and microscopic characters could possibly yield a one-species concept of these brown-spot causal agents. Not only were there several distinctive, segregable morphogroups, plus a scattering of unique isolates, but also there were some geographic discontinuities for groups from different citrus-growing regions.

A ready conclusion, based solely on morphologic criteria, is that the pathologic condition generally known as "*Alternaria* brown spot of mandarins" can be caused by several quite different species of *Alternaria*.

It is noteworthy, again, that no isolate among these 80-90 from lesions of tangelo and related cultivars, is identifiable with typical *Alternaria alternata*. It is remarkable, likewise, that the Whiteside isolate PR320 (EGS 39-181, Kohmoto AC320, ATCC 38963), used in characterizing the pathology of "the tangerine pathotype of *Alternaria alternata*" (Kohmoto et al., 1991) not only cannot be identified culturally or microscopically with typical *A. alternata* but indeed is morphologically unique in the present context, without a single identical isolate within the set of 80-90 pathogenic isolates from tangerine-related cultivars.

An observation made during initial comparisons is that numerous isolates from Israel, South Africa, and Turkey are similar among themselves, but that they differ markedly from those originating in Colombia and Florida. Some of the distinctive groups of isolates from the several study regions have as many as a dozen or more representatives, other groups 3-6 strains, and a few only one or two decidedly different isolates.

It would be an almost endless, predictably thankless task to describe and illustrate the definable stable diversity seen here under controlled culture conditions. My decision has been to select for description those taxa represented by five or more isolates, along with examples of unique isolates. Coverage is for nearly 60% of the available material. This leaves the less well-represented small groups and less distinctive individual isolates in taxonomic limbo for the present but does not substantially degrade the thesis that many different species of *Alternaria* can induce the brown spot pathology of tangerine-related cultivars.

Descriptions of three major-group and five unique isolates from among the set derived from Minneola tangelo and related cultivars are presented in the following order:

228. *A. tangelonis*; the major group of 20+ isolates from tangelo; Colombia and Florida.
229. *A. citriarbusti*; a group of 4+ isolates; Florida.
230. *A. turkisafria*; the major group of 15 isolates from tangelo; Israel, South Africa, Turkey.
231. *A. toxicogenica*; unique in this set (Whiteside PR320); from Dancy tangerine; Florida.
232. *A. colombiana*; a single isolate; tangelo; Colombia.
233. *A. perangusta*; a single isolate; tangelo; Turkey.
234. *A. interrupta*; a single isolate; tangelo; Israel.
235. *A. dumosa*; a single isolate; tangelo; Israel.

* * * * *

228. **Alternaria tangelonis** Simmons, sp. nov. Fig. 157

Etym.: hort., tangelo (L., f., gen.), *Citrus* x tangelo (*Citrus reticulata* Blanco x *C. paradisi* Macf.)

Ex cultura in agaro Hay descripta. Coloniae modice concentrice zonatae, dense conidiogenae. Conidiophora primigena plerumque 40-60 x 4-5μm. Catenae conidiorum simplicis, constatae ex conidiis 4-8(-10). Conidia typice et diagnostice angustiovoidea, extensione apicali longiangusta (conidiophoro secundario), 50-90 x 8-13μm, 5-7 transverse et 1-3 longitudinaliter septata, subhyalina ad dilute *brunnea, laevia. Habitatio typi in laesione Citrus paradisi Macfad. x C. reticulata Blanco cv. Minneola, in arbusto citreo, Lake Alfred, Florida, U.S.A. Typus: partes ex cultura EGS 45-114 (isol. EV-MIL-2s ex CREC, Lake Alfred, Florida; accept. viii. 1997) desiccatae et in* ***BPI*** *(holotypus) et saltem in* ***IMI*** *(isotypus) conservandae.*

Colonies on all media used have concentric rings of growth and sporulation. On PCA, rings of relatively short to moderately long conidiophores (after light exposure) alternate with rings of cottony aerial growth which is composed of interwoven fascicles of long, semierect hyphae that function as primary conidiophores. On V-8, ring formation is less pronounced because most of the colony is composed of the dense cottony growth of intertwined sporulating fascicles. On Hay, dominant rings (light exposure) consist of close stands of relatively short primary conidiophores, with alternate rings containing only a few tall, uncrowded sporulating hyphae.

On PCA, **primary conidiophores** in light-induced rings are simple, to about 60-120 x 4-5µm, or commonly 2-branched, to ca. 240µm in length, with each suberect branch ca. 120 x 4-5µm; aerial fascicles (dark phase growth) are 1.0mm or more long, with several lateral and terminal branches often 60+µm in length. Most of the primary conidiophores on Hay (light phase) are 40-60 x 4-5µm. The long fascicles of aerial hyphae that dominate V-8 colonies have subterminal and terminal branches ca. 80-125 x 4-5µm.

Conidia are borne in simple chains that only occasionally have 1-2 short lateral branches. On PCA, chains have 6(-8) conidia in light phase, 1-4 on aerial branches; on Hay, 4-8(-10) in light phase, 1-4 on aerial branches; on V-8, 4-5 in light, 1-3 on aerial branches.

Conidium shape and size vary considerably but characteristically in sporulation on the three different media. Sporulation is very distinctive on Hay; the species is typified, therefore, on cultures produced on this medium. Sporulation on V-8 also is distinctive; when coupled with that from Hay, the diagnosis of the species among citrus isolates of the present set becomes firm.

The most striking, sometimes dominant **conidia in Hay** cultures have a narrow-ovoid base with a long narrow (but not filiform), tapering apical extension that functions as a secondary conidiophore within the chain. Other conidia, ovoid with only a very short 1-2-cell secondary conidiophore,

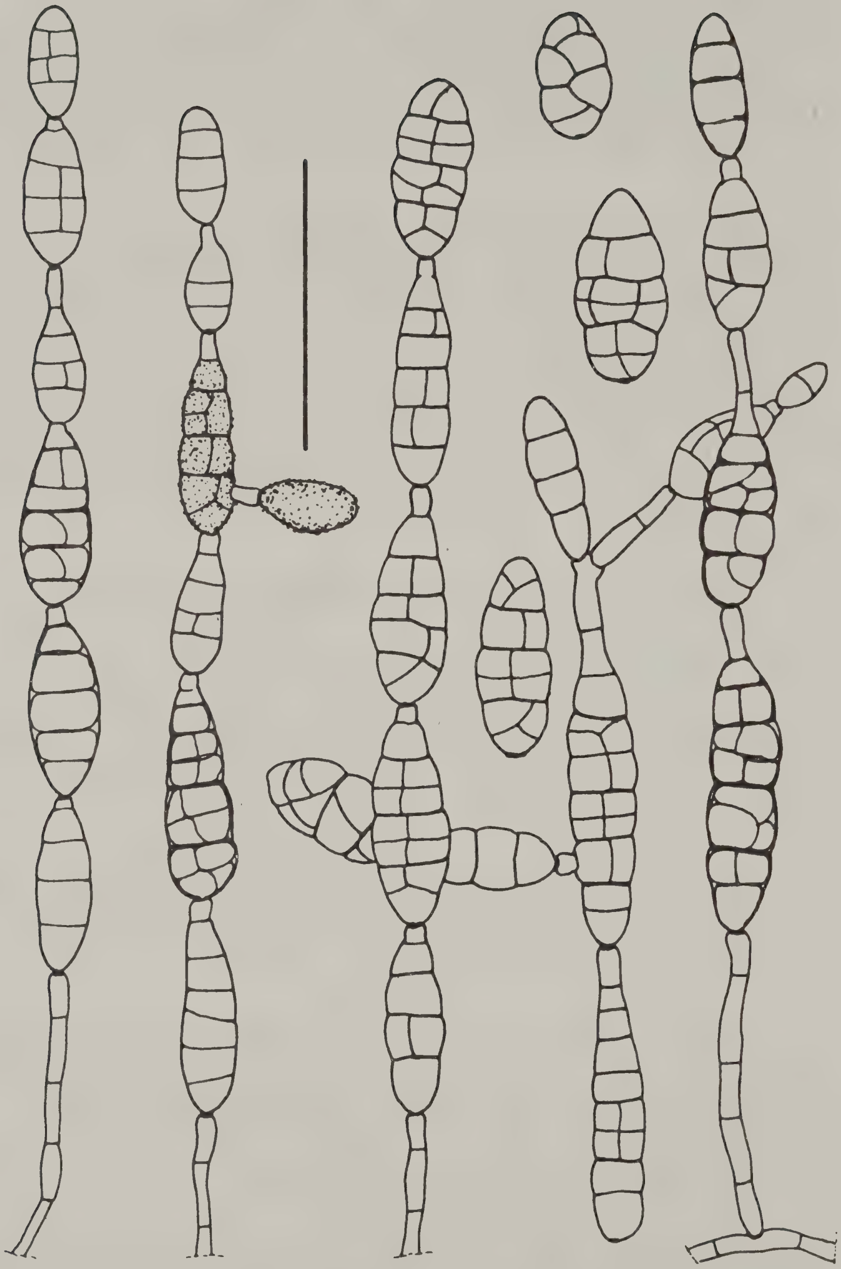

Fig. 157. *Alternaria tangelonis*: conidia and conidiophores ex type isolate EGS 45-114. At left, 6d. on PCA; at right, 6d. on V-8. Bar = 50μm.

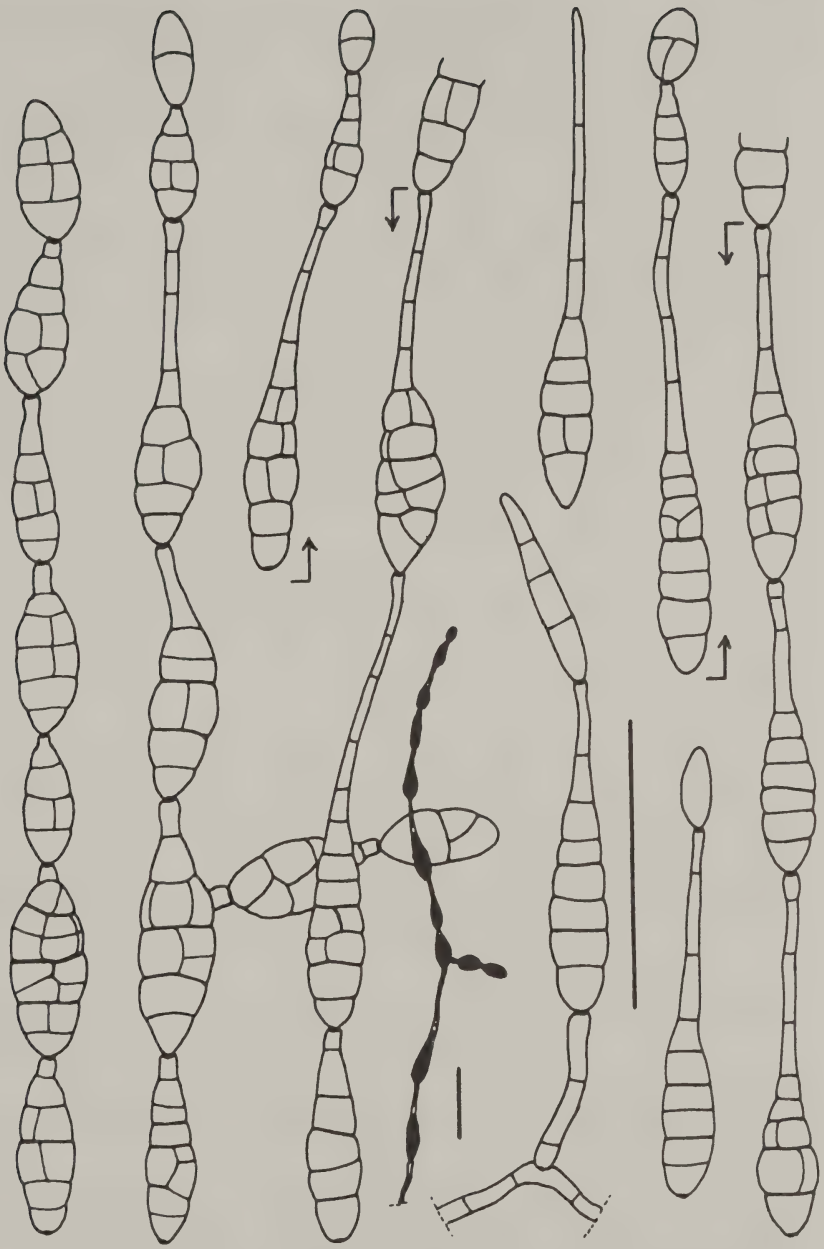

Fig. 157 contd. *Alternaria tangelonis*: conidia, conidiophores, habit ex type isolate EGS 45-114; 6d. on Hay. Bars = 50μm.

may occur in the same chains as do those with long apical extensions; or they may occur in simple chains of conidia with only short apical conidiophores. The diagnostic long-narrow conidia are 50-90 x 8-13μm, with 5-7 transepta in the ovoid base and 1 longiseptum in each of 1-3 of the transverse segments. Conidia from Hay are smooth-walled, subhyaline to dilute tan.

Conidia in the relatively short chains that are produced on **V-8** are basically narrow- to broad-ellipsoid. They are separated in the chain by short 1-2-cell secondary conidiophores ca. 3-12 x 3μm. Because the chains are short, approximately 20-30% of the conidia are terminal and thus lack an apical secondary conidiophore. These terminal conidia characteristically are broad-ellipsoid, strongly constricted at 3-4 major transepta, ca. 30-14μm, with additional transverse or oblique septa in each major segment. These distinctive terminal conidia, constituting a conspicuous portion of the conidial population, are a strong diagnostic character when considered in conjunction with the very long narrow conidia produced in abundance on Hay agar. Conidium bodies of spores produced on V-8 may reach a size of 30-50 x 11-14μm; they are medium brown, are mostly smooth with occasional minor punctulation, and have 3-4 major transepta plus secondary transverse, oblique and longitudinal septa in most of the major segments.

Conidia produced on **PCA**, though in short chains, are not distinctive enough to be diagnostic in the present context of citrus isolates. Conidia are ovoid, usually have a short apical secondary conidiophore (3-8 x 3μm), are 20-35 x 8-12μm, have 3-6 major transepta plus secondary transverse, oblique and longitudinal septa in each major segment, are medium brown and mostly smooth but with conspicuously strong, punctate ornamentation on largest, oldest conidia.

Alternaria tangelonis represents the largest group of identifiable isolates in the study set of 54 derived from Minneola tangelo lesions sampled in the Florida and Colombia study areas. At least 25% of the isolates examined from these two regions are of this species. *A. tangelonis* is not present among the 30 isolates studied from Minneola tangelo sources in Israel, South Africa, and Turkey.

A partial list (all from Minneola tangelo lesions) includes isolates from Colombia (1994: EGS 41-168/-169/-170/-175/-177; from Colombia (1996): EGS 45-014/-015/-016; from Florida, U.S.A. (1997): EGS 45-114/-115/-116/-120/-121/-122/-125; from Florida (1998): EGS 46-139/-142/-143/-147/-148/-149. See Appendix for original source information.

229. **Alternaria citriarbusti** Simmons, sp. nov. Fig. 158

Etym.: L. (gen.), *Citrus* + *arbustum* = of citrus grove

Ex cultura in agaro V-8 descripta. Coloniae concentrice zonatae, dense conidiogenae. Conidiophora primigena ad 200 x 5µm (in luce), in fasciculis ad 60-800µm longis (in statu obscuro). Catenae conidiorum constatae ex conidiis 2-6; conidia plerumque solitaria. Conidia longiellipsoidea, ad ca. 50-80 x 10-12µm, autem conidia terminalis vulgo ca. 40-50 x 9-12µm; conidiophoris secundariis 3-5 x 4µm vel 25-35 x 2-3µm; 7-9(-11) transverse, 1-4 longitudinaliter septata; modice sordide brunnea, punctulata vel punctata. Habitatio typi in laesione Citrus paradisi Macfad. x C. reticulata Blanco cv. Minneola, in arbusto citreo "Shinn Grove," Florida. Typus: partes ex cultura EGS 46-141 (isol. SH-MIL-15s ex CREC, Lake Alfred, Florida; accept. ii.1998) desiccatae et in BPI (holotypus) et saltem in IMI (isotypus) conservandae.

Colonies on all media used have concentric rings of growth and sporulation, produced in response to the influence of the alternating light/dark cycle. In this species it is characteristic for most of the primary conidiophores to be relatively long. On PCA, rings of an open tangle of conidiophores (light exposure) up to ca. 200 x 5µm alternate with rings of intertwined fascicles of conidiophores (dark phase) up to 600-800µm. On Hay, production of primary conidiophores is more open than on PCA, often with 2-3 conidiophores arising in a group from the agar surface but not in close fascicles. On V-8, primary conidiophore production is much like that on PCA, but with the light-phase stand of conidiophores dense and obscured by a dense, black mass of conidia and the aerial mass of tall, branching, intertwined conidiophores much more dense than that on PCA. Conidiophores may be unbranched on all media, but very commonly produce 1-3 lateral branches at intervals. All solitary conidiophores and all branches of conidiophore fascicles support terminal solitary chains of conidia.

Conidia are borne in simple short chains that usually are unbranched. Chains (PCA) have 5-8 conidia (light phase), 1-4 on aerial branches; (Hay) up to ca. 8 conidia (light phase rings), 1-3 on short aerial branches; (V-8) 1-6 conidia (light phase), 1-3 on aerial branches.

The characterizing shape of large numbers of **conidia** on all media is long-ellipsoid or narrow-ovoid, occasionally with a moderately long secondary conidiophore as an apical extension, but usually with a relatively short 1-cell second-

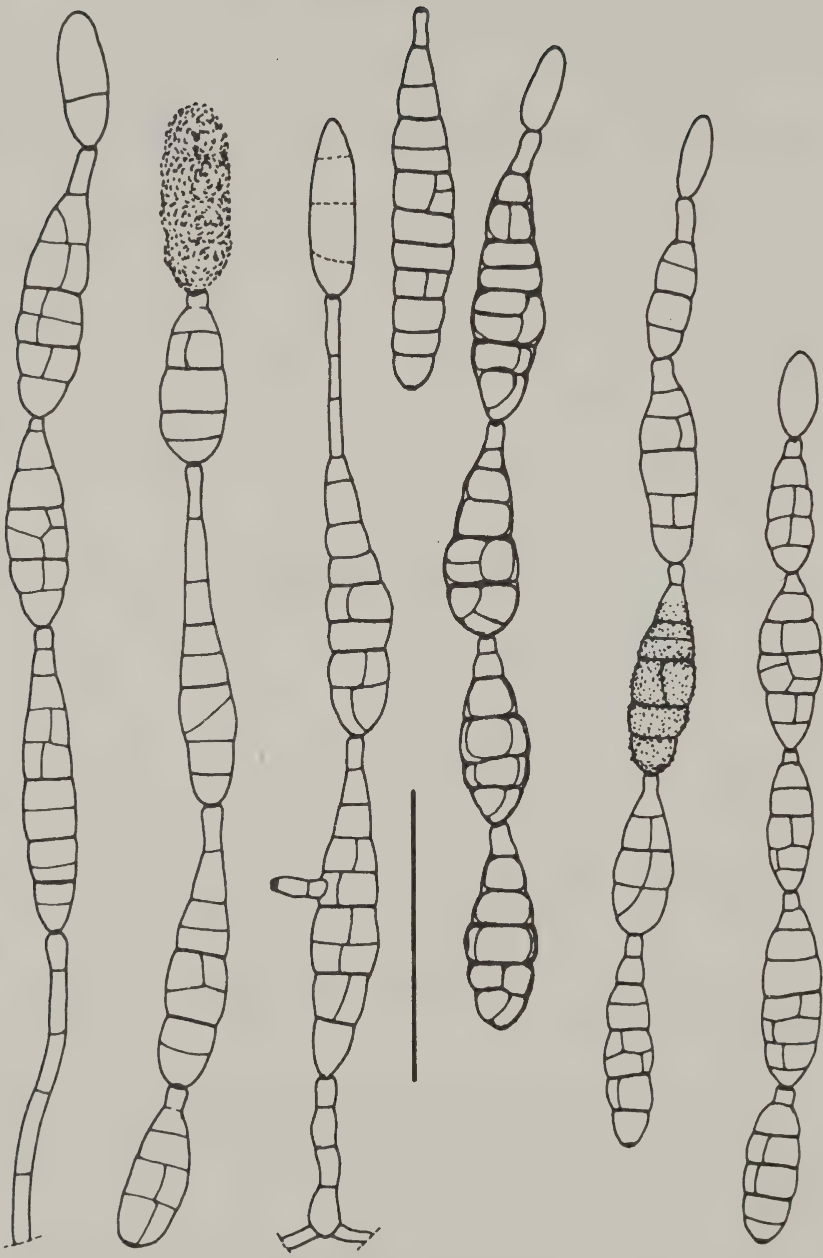

Fig. 158. *Alternaria citriarbusti*: conidia and conidiophores ex type isolate EGS 46-141; 6d. on PCA. Bar = 50μm.

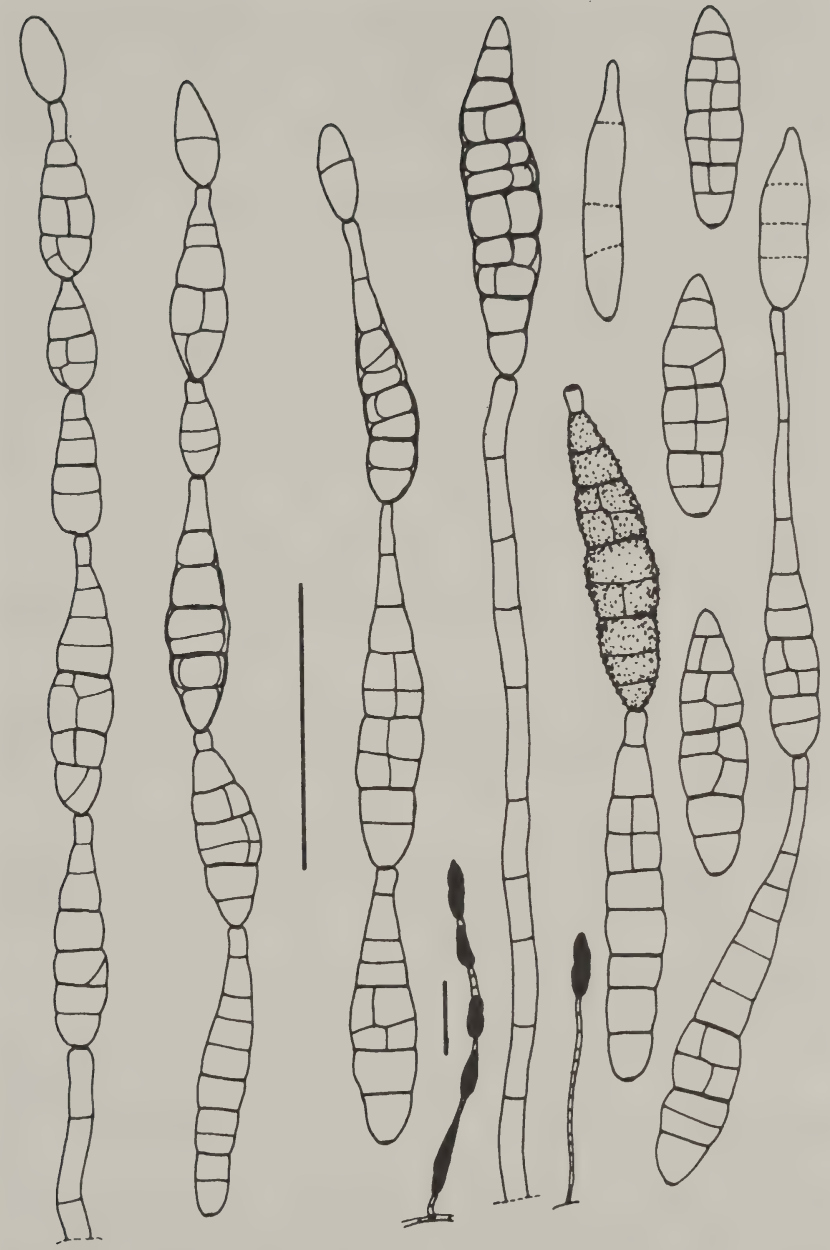

Fig. 158 contd. *Alternaria citriarbusti*: conidia, conidiophores, habit ex type isolate EGS 46-141. At left, 6d. on Hay; at right, 6d. on V-8. Bars = 50μm.

ary conidiophore. On **PCA, chains** often contain conidia that are ovoid, ca. 30 x 12μm; but the dominant pattern is narrow-ovoid, 30-60 x 8-12μm, with 6-11 transepta and 1 longiseptum in a few of the transverse segments. On **Hay, chains** include both narrow-ellipsoid and moderately broad-ovoid conidiam reaching 35-40 x 8μm (narrow components) and ca. 25-30 x 10-11μm (ovoid). On V-8, enormous numbers of conidia with a distinctive long-ellipsoid shape are produced; the species is typified, therefore, on cultures produced on this medium.

The narrowly ellipsoid **conidia** produced on **V-8** frequently remain solitary at the conidiophore tip or, more commonly, develop in short chains of up to 6 spores. Conidia reach a dominant range of ca. 50-80 x 10-12μm; distinctive terminal conidia (secondary conidiophore lacking) may reach the maximum size but more commonly are ca. 40-50 x 9-12μm. Apical secondary conidiophores may be only a 3-5 x 4μm single cell or an elongate but not filiform extension up to 25-35 x 2-3μm. Conidium bodies are a dull medium brown with darker walls and major septa; are usually evenly punctulate to punctate; and have as many as 7-9(-11) transepta, of which at least the near-median one is conspicuously dark and constricting, and 1-2 longitudinal septa in a few of the transverse segments. Differing from many species whose conidia are mostly distoseptate at 7d., V-8 conidia of *A. citriarbusti* nearly always have one and very commonly 4-5 dark, slightly thickened, noticeably constricting eusepta plus one longitudinal euseptum in a few segments and a balance of secondary transverse and longitudinal distosepta in remaining segments.

The isolates of *A. citriarbusti* originated at a single locale of Minneola tangelo growth known to the Lake Alfred citrus center staff as Shinn Grove. Representative isolates, which reached me in February 1998 (all from leaf lesions), include at least EGS 46-136, 46-140, 46-141, and 46-150. See Appendix for original source information.

230. **Alternaria turkisafria** Simmons, sp. nov. Fig. 159

Etym.: mnemonic, from Turkey, Israel, South Africa; indicating the presently known geographic distribution of this taxon.

Ex cultura in agaro V-8 descripta. Coloniae fortiter concentrice zonata. Conidiophora brevia (in luce) vel ad 50-100μm (in statu obscuro). Catenae principales conidiorum constatae ex conidiis 8-20. Conidia longiangusta, 30-55 x 6-8(-10)μm, conidiophoris secundariis 3-10(-20) x 2-3μm, 8-10(-14) transverse, raro 1-3 longitudinaliter septata; modice *luteobrunnea, laevia vel punctulata vel punctata. Habitatio typi in laesione Citrus paradisi Macfad. x C. reticulata Blanco cv. Minneola, in arbusto citreo, Kuzucuoglu, Turkey. Typus: partes ex cultura EGS 44-166 (Y. Canihos, YC 27, v.1996 isol.) desiccatae et in* ***BPI*** *(holotypus) et saltem in* ***IMI*** *(isotypus) conservandae.*

Colonies on all media used have concentric rings of growth and sporulation. These are most pronounced on V-8, where black rings (light exposure) of a dense growth of primary conidiophores and enormous numbers of conidia alternate with taller rings (dark phase) of a dense cottony growth of aerial conidiophores and conidial chains. On PCA and Hay, rings are less evident because conidiophores of both light exposed and dark phases scarcely differ in length and branching and do not develop in the dark phase as a distinct ring of tightly interwoven sporulation elements (as occurs on V-8).

The 50X aspect of PCA and V-8 colonies is of a tangle of variously curved, very long, narrow chains of conidia. The 50X picture of Hay colonies is similar except that conidiophores are uncrowded and their chains less intimately tangled.

Individual primary conidiophores on PCA and Hay are relatively short (ca. 30-60 x 4μm), arising either directly from the agar surface or as lateral branches of weak, somewhat creeping surface and aerial hyphae that are obscured by the abundant, tangled conidial chains. On V-8, individual primary conidiophores also may be relatively short in light-exposed areas but somewhat longer (50-100μm) when they are produced in the dense, cottony aerial hyphal growth of the dark-phase rings.

Conidia on all media are borne in long simple or branched chains. Chains (PCA) have 8-20 conidia, plus as many as 4-10 on branches; (Hay) up to 15, with fewer in branches; (V-8) up to 20 in the initial chain, with fewer (commonly ca. 5) on aerial branches.

Conidia characteristically are long-ellipsoid or long-ovoid on all media. Shorter ovoid units occur, especially on Hay, but the population is strongly dominated by the habit of long-narrow spores in very long chains. This character is accentuated in sporulation on V-8 agar; cultures on V-8 are used in typification of the species.

Conidia produced on **PCA** are narrow-ovoid to long-ovoid or long-ellipsoid, 20-50 x 6-8μm, have small 1-cell secondary

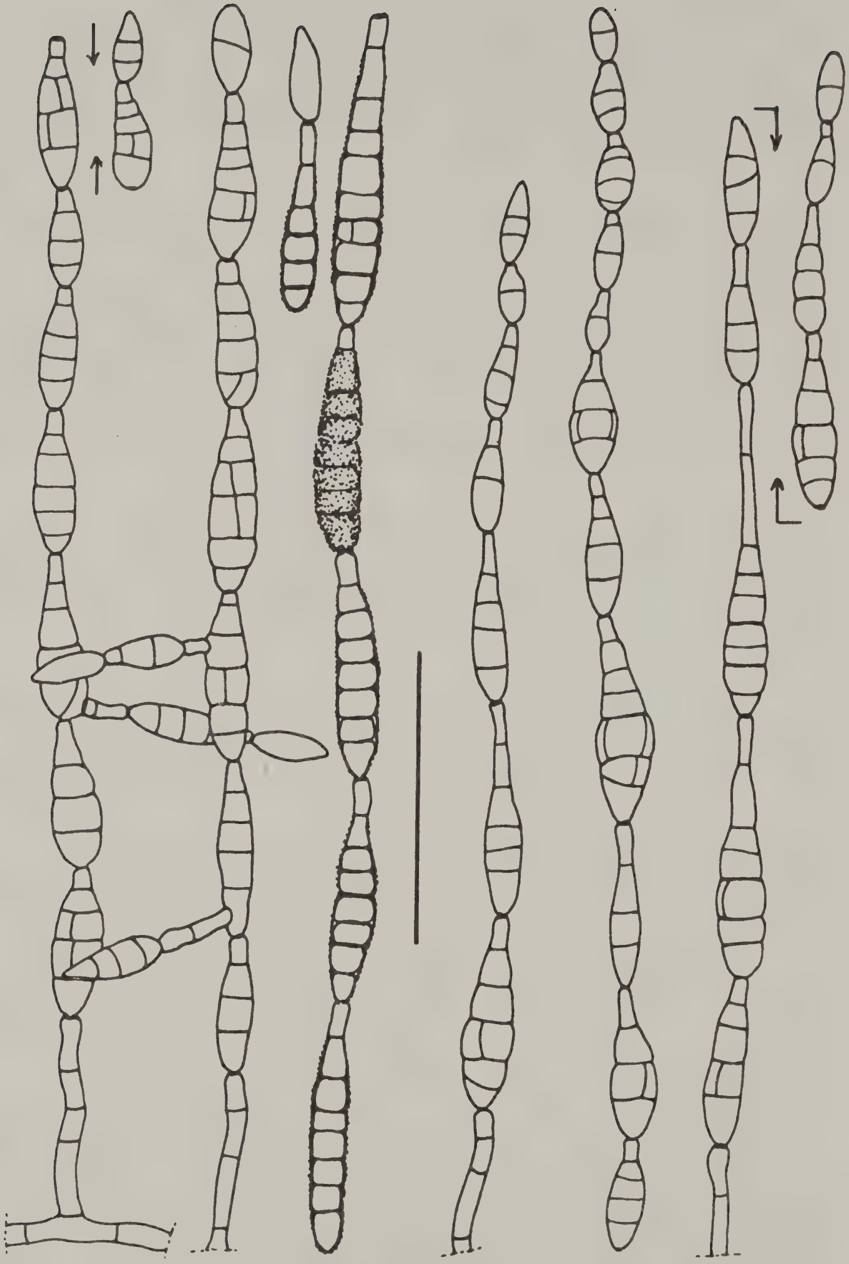

Fig. 159. *Alternaria turkisafria*: conidia and conidiophores ex type isolate EGS 44-166. At left, 5d. on PCA; at right, 5d. on Hay. Bar = 50µm.

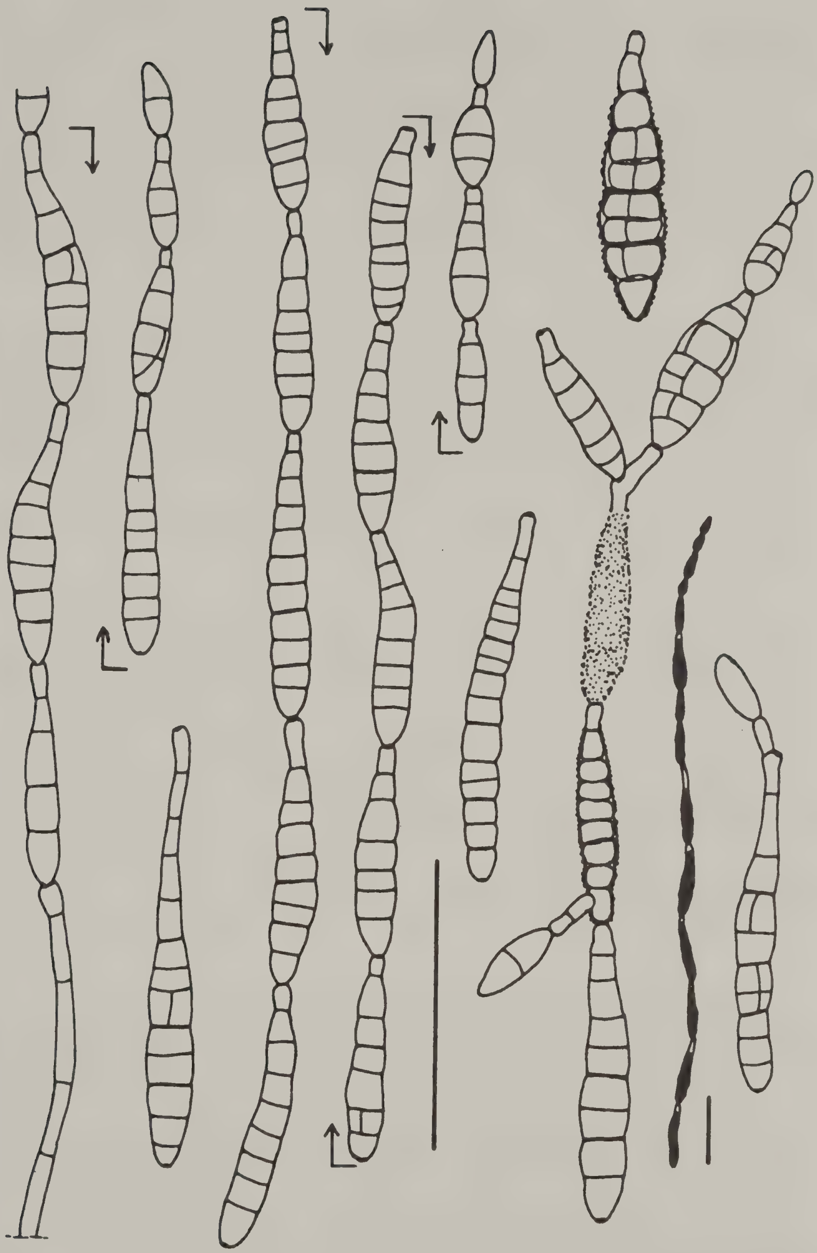

Fig. 159 contd. *Alternaria turkisafria*: conidia, conidiophores, habit ex type isolate EGS 44-166; 5d. on V-8. Bars = 50μm.

conidiophores 3-5 x 2-3μm, 3-8 transepta and usually no longisepta or, at most, 1 longiseptum in 1-2 transverse segments, are a medium yellow-brown and evenly punctate overall. Conidia produced on Hay also are predominantly elongate, although considerable numbers of short-ovoid units also are present. Conidium bodies in a single chain may vary between lengths of 10-35μm. Largest conidia reach a range of ca. 25-35 x 8-11μm and have apical secondary conidiophores ca. 3-10 x 2-3μm; long narrow secondary conidiophores up to 15-40 x 2-3μm commonly are found on 1-2 conidia in some chains.

Conidia produced on **V-8** are predominantly long and narrow; broader ovoid and ellipsoid spores are present but in relatively small numbers. Conidium bodies are 30-55 x 6-8(-10)μm, have a 1(-2)-cell apical secondary conidiophore 3-10(-20) x 2-3μm, up to 8-10(-14) transepta and mostly no longisepta or, at most, 1 longiseptum in 1-3 segments (sometimes 1(-2) longisepta in 4-5 of the transverse segments of the broader ovoid conidia), are a medium yellow-brown and smooth to punctulate to punctate.

Alternaria turkisafria is represented by at least 15 of the 30 isolates studied from Minneola lesions from sampling locations in Turkey, Israel, and South Africa. The species is not present among isolates examined from Florida and Colombia. A record list of the 15 isolates includes Turkey: EGS 44-159/-161/-162-/-163 (unstable) /-164/-166/-167; Israel: 45-003/-004/-005 (unstable) /-006 (probable); South Africa: 45-056/-057/-058/-059 (unstable). See Appendix for original source information.

231. **Alternaria toxicogenica** Simmons, sp. nov. Fig. 160

Etym.: L + ISV, *toxicum* + *-genic*, toxin-producing

*Ex cultura in agaro V-8 descripta. Coloniae fortiter concentrice zonatae. Conidiophora brevia (in luce) vel ad 500-600μm (in statu obscuro). Catenae conidiorum constatae ex conidiis 2-6 (raro -10); conidia plerumque solitaria. Conidia robusta, late ovoidea vel late ellipsoidea, 35-70 x 15-18μm, conidiophoris secundariis 3-20 x 2-3μm, 7-9 transverse et modice irregulariter longitudinaliter vel oblique septata; translucide dilute avellanea, septis fuscioribus, laevia vel punctulata vel fortiter punctata. Habitatio typi in laesione Citrus reticulata Blanco cv. Dancy, in arbusto citreo, Wauchula, Florida, U.S.A. Typus: partes ex cultura EGS 39-181 (J. O. Whiteside PR 320, vi.vi.1975 isol.) desiccatae et in **BPI** (holotypus) et saltem in **IMI** (isotypus) conservandae.*

Colonies on V-8 exhibit concentric rings of growth and sporulation, with dense rings (light exposed) of relatively short conidiophores and masses of conidia alternating (dark phase) with taller, crowded stands of branching primary conidiophores and abundant conidial chains. On PCA and Hay, there is little distinction between light/dark-phase areas; short and long primary conidiophores are not crowded, are totally sporogenous, and thus do not present well-defined alternating rings of sporulation.

Individual primary conidiophores on PCA are 50-100 x 4-5μm, mostly shorter on Hay. On V-8, primary conidiophores (light exposed) are short as on PCA and Hay; those in dark-phase areas are considerably longer, to 500-600μm, with branches in the 50-100μm range. Simple conidiophores on all media commonly become geniculate once or twice near the apex, and generate 2-3 individual chains of conidia.

Chains of conidia are short on all media, 2-6 (rarely -10); conidiophore geniculations and branches very often produce only a single spore. Although most chains are unbranched, at least in early colony growth, it is common and characteristic for individual conidia to produce 1-2 lateral secondary conidiophores and short chains of 2-3 units. This internal branching coupled with multiple chain formation on multi-geniculate conidiophores can generate small branching clumps of 15-20+ conidia.

The initial impression of slide preparations from all media is of relatively large, broad-ovoid and broad-ellipsoid conidia with well-constricted, sometimes irregular septation. There is always a conspicuously high proportion of conidia with a blunt or broadly rounded apex, these being conidia that were noncatenate or that were terminal in the typically short chains.

On **PCA, conidia** in young chains are long-ovoid, ca. 30-50 x 11-16μm; in older chains, broad-ellipsoid, ca. 25-45 x 15-20μm; have 1-2-cell secondary conidiophores ca. 3-12 x 3μm; usually have a median constricting septum and 2-6 additional transepta, with internal secondary oblique and longitudinal septa somewhat irregularly inserted; are a medium dull gold-brown and smooth to punctulate or sometimes strongly punctate.

On **Hay, conidia** are long-ovoid, ca. 30-50 x 12-15μm; have short 1-cell secondary conidiophores; have 5-8 transepta, a few of which are constricting, and several irregularly inserted oblique and longitudinal secondary septa; are subhyaline to medium tan (with septa contrastingly darker)

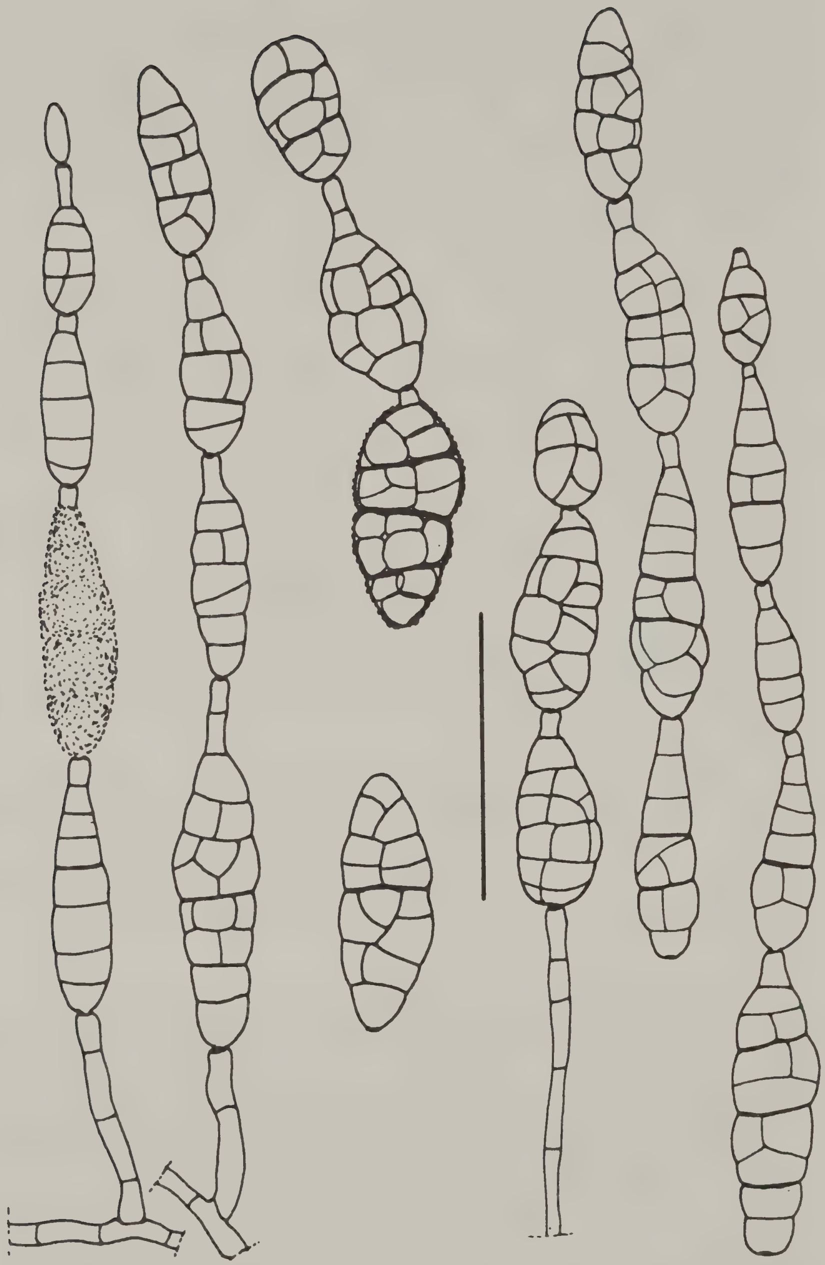

Fig. 160. *Alternaria toxicogenica*: conidia and conidiophores ex type isolate EGS 39-181. At left, 6d. on PCA; at right, 6d.on Hay. Bar = 50μm.

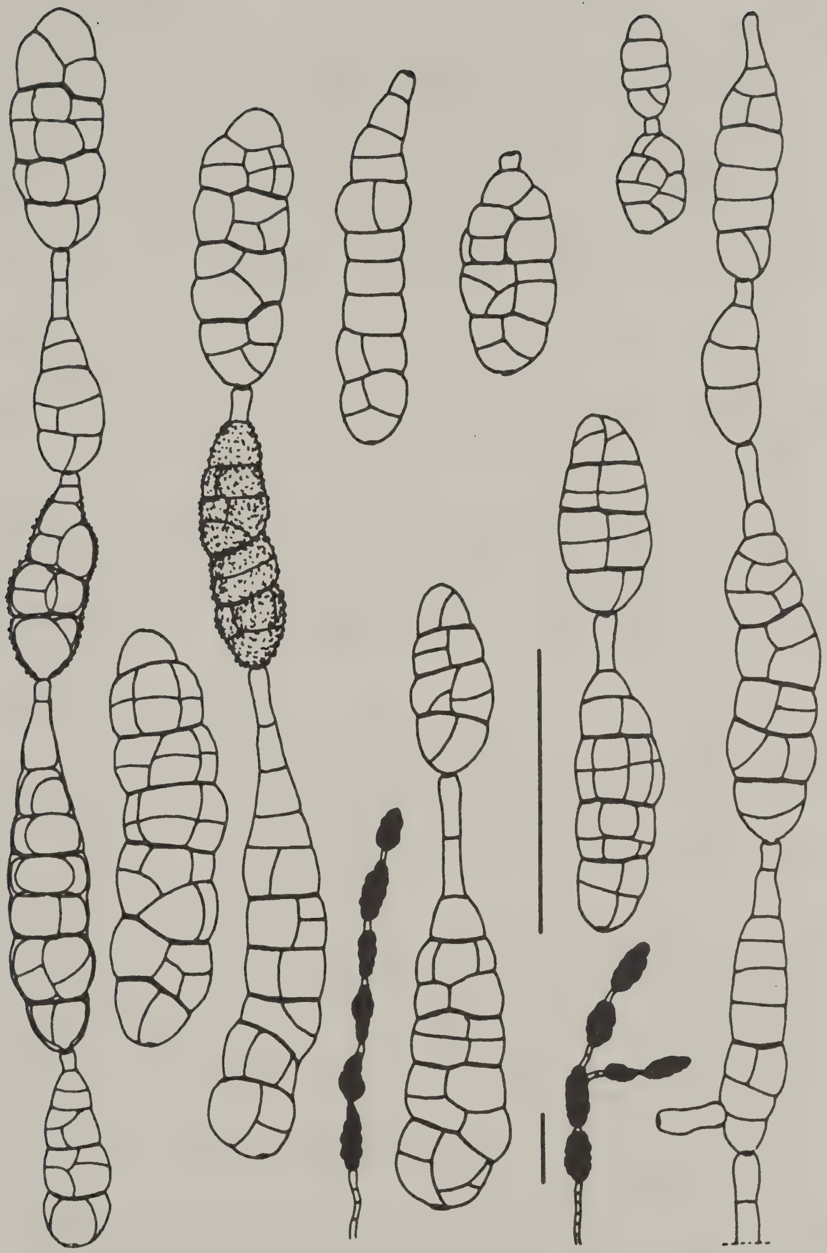

Fig. 160 contd. *Alternaria toxicogenica*: conidia, conidiophores, habit ex type isolate EGS 39-181; 6d. on V-8. Bars = 50μm.

and smooth to punctulate.

On **V-8**, which is used in typification of the species, **conidia** are robust, broad-ovoid or broad-ellipsoid, 35-70 x 15-18μm; have 1(-2)-cell secondary conidiophores 3-20 x 2-3μm, which arise abruptly from the blunt apex of the conidium; usually have 7-9 transverse or oblique septa, of which 3-5 are primary and constricting, and several oblique and longitudinal secondary septa; are a translucent tan (with septa contrastingly darker) and smooth to punctulate to strongly punctate.

Alternaria toxicogenica is known to me only through isolate EGS 39-181 (Whiteside PR320), which was received in September 1990 from J. O. Whiteside as being "from Dancy tangerine leaves with *Alternaria* brown spot, Wauchula, FL, June 6, 1975...pathogenicity recently verified."

A Whiteside paper (1976b) recorded and discussed an *Alternaria*-induced brown spot disease of Dancy tangerine as new in Florida. Whiteside worked with filtrates from cultures of the isolate and used the term "host-specific toxic filtrates" in discussing his experimental results. He did not cite a strain number for the isolate used.

The Whiteside isolate PR320 (as AC-320) was the subject organism in a study of host-specific toxins from "the tangerine pathotype of *Alternaria alternata*" (Kohmoto et al. 1991). The isolate appeared again (as ATCC 38963 = PR320) in a phylogenetic analysis of HST-producing *Alternaria* isolates, with the listing "*A. citri* (the tangerine pathotype)" (Kusaba and Tsuge 1995).

This Whiteside isolate is morphologically unique among the few isolates at hand from tangerine (Florida 1990, 1996) and has no morphological equivalent among the 70+ brown spot isolates from Minneola tangelo in this study. Within the context of moderately small-spored *Alternaria* species, it is inconceivable that any system of classification-directed comparison of cultures, sporulation pattern, or conidium morphology under controlled conditions would lead to identification of *A. toxicogenica* with either *A. alternata* (Simmons 1967, 1992; Simmons and Roberts 1993) or typical *A. citri* (Simmons 1990).

232. **Alternaria colombiana** Simmons, sp. nov. Fig. 161

Etym.: geogr., Colombia, country of origin

Ex cultura in agaro V-8 descripta. Coloniae fortiter *concentrice zonatae. Conidiophora 150-200 x 4µm (in luce) vel ad 1mm (plus minusve) x 4-5µm (in statu obscuro). Catenae conidiorum constatae ex conidiis 2-5, vel pro maxima parte non nisi 1-2. Conidia longe angustiovoidea vel angusti-ellipsoidea, 35-75 x 10-12µm (in coloniis juvenibus) aut ovoidea vel ellipsoidea 32-50 x 17-21µm (in coloniis magis veteribus), conidiophoris secundariis 20-125 x 3-4µm, 7-9 transverse et 1-2 longitudinaliter septata, dilute luteo-brunnea, laevia vel punctulata. Habitatio typi in laesione fructus Citrus paradisi Macfad. x C. reticulata Blanco cv. Minneola, in arbusto citreo, Chinchiná, Colombia. Typus: partes ex cultura EGS 45-017 (B. L. Castro C., C-7; accept. xi.1996) desiccatae et in BPI (holotypus) et saltem in IMI (isotypus) conservandae.*

Colonies on V-8 exhibit concentric rings of growth and sporulation, with dense rings (light exposed) of relatively short conidiophores and dark masses of conidia, alternating (dark phase) but also intermixed with very tall, crowded, simple or branching primary conidiophores with a solitary apical conidium or short chain of conidia. On PCA and Hay, there is little distinction between light- and dark-phase areas, where moderately to very long conidiophores (PCA) and relatively short conidiophores (Hay) are not crowded, always are sporogenous, and thus do not present a growth pattern of alternating rings.

On **PCA, primary conidiophores** are up to 80-350 x 4-5µm; on **Hay**, 100-150+ x 3-4µm. They may remain simple, but very often branch and occasionally extend beyond conidiogenous geniculations. On **V-8, primary conidiophores** may be ca. 150-200 x 4µm, but more conspicuously are very long, 1 mm or more x 4-5µm, with ascending branches and geniculate extensions as well as multiple branches near the apex.

Solitary **conidia** at the tips of conidiophores and their branches are abundant on all media. Typically on PCA, however, **chains** of 2-5 conidia, often separated by very long secondary conidiophores, are produced. Chains may be simple, but branching by means of long geniculate extensions is very common.

On **Hay**, the sporulation pattern is the same as that on PCA except that the stand of primary conidiophores is much more open and the progression of conidiophore and chain development more easily examined at 50X.

On **V-8**, production of short chains is common, but the dominant 50X picture by far is of the very long primary conidiophores, with their geniculate extensions and branches,

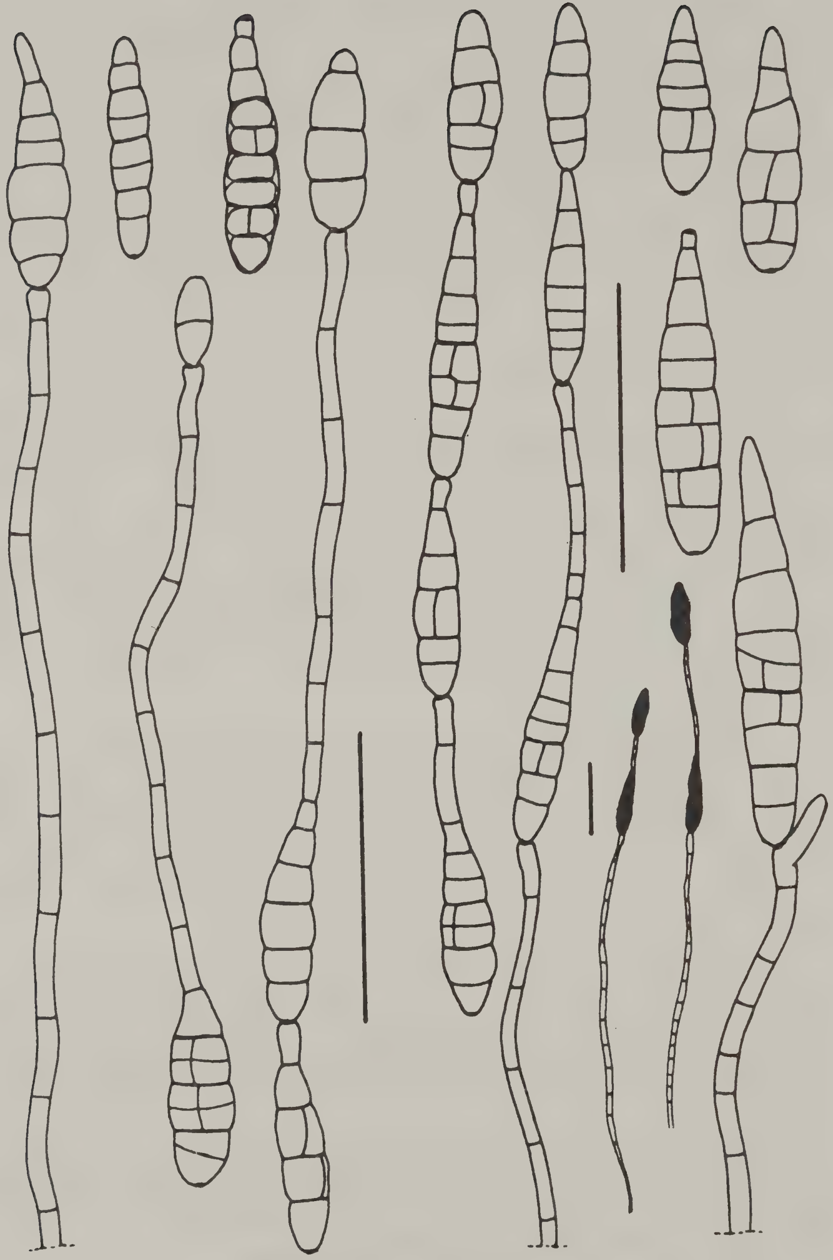

Fig. 161. *Alternaria colombiana*: conidia, conidiophores, habit ex type isolate EGS 45-017. At left, 4d. on PCA; at center, 4d. on Hay; at right, 4d. on V-8. Bars = 50μm.

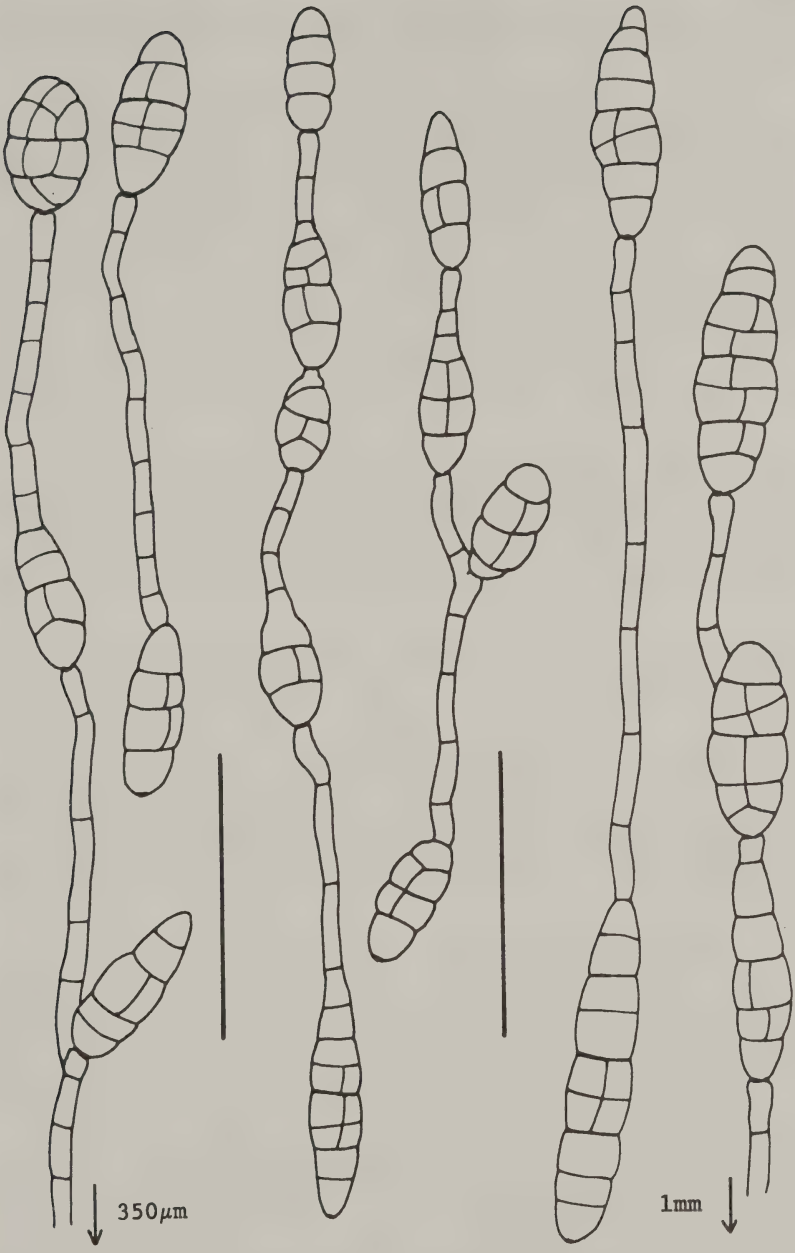

Fig. 161 contd. *Alternaria colombiana*: conidia and conidiophores ex type isolate EGS 45-017. At left, 10d. on PCA; at center, 10d. on Hay; at right, 10d. on V-8. Bars = 50µm.

at the tips of which only a single conidium or short chain of two is produced.

On **PCA, conidium** bodies are narrow-ovoid, 30-40 x 7-10μm; sometimes have a 1-cell secondary conidiophore ca. 3-5 x 3μm, but usually are separated by very long, multiseptate secondary conidiophores up to ca. 100-120 x 3-4μm; have 3-7 transepta and 1 longiseptum in 1-2 of the transverse segments; are dilute yellow-tan and smooth to evenly punctulate. As the colony ages to 7-10 days, new conidia are short-ovoid, commonly in a size near 25-30 x 16μm; have correspondingly fewer septa; and are a darker medium brown. Conidia are primarily distoseptate in colonies of all ages, with a median euseptum distinct and somewhat constricting in most conidia, and sometimes 1-2 additional primary transverse eusepta.

On **Hay, conidia** are narrow-ovoid or narrow-ellipsoid, 30-50 x 6-8μm; have a short 1-cell secondary conidiophore ca. 5 x 3μm or relatively long, multiseptate secondary conidiophores up to ca. 40-50μm long; have 5-8 transepta and 1 longiseptum in 1-2 of the transverse segments; and are dilute yellow-tan and smooth.

On **V-8**, which is used in typification of the species, **conidia** are long-narrow-ovoid or narrow-ellipsoid, 35-75 x 10-12μm in 5-7d colonies, tending toward broader ovoid and ellipsoid, 32-50 x 17-21μm in colonies 7-10d old; commonly have multiseptate secondary conidiophores 20-125 x 3-4μm, have a pigmented, constricting, transverse median euseptum in most conidia and sometimes 1-2 additional scarcely-constricting primary transverse eusepta plus 5-6 secondary distosepta, and 1 longiseptum in 1-2 of the transverse segments; and are dilute yellow-tan and smooth to punctulate.

The sporulation habit of *A. colombiana* immediately invites comparison with *A. pseudorostrata* Simmons (1996, *Alt.T&V* 147) in that very long secondary conidiophores inserted in short chains of beakless conidia are characteristic of both species. Sporulation elements of *A. pseudorostrata* appear conspicuously more robust at 50X and higher magnifications than those of *A. colombiana*. Specifically, *A. pseudorostrata* (on PCA): conidiophores 150-400 x 6-8μm, conidia 55-90 x 14-20μm; *A. colombiana* (on PCA): conidiophores 80-350 x 4-5μm, conidia 30-40 x 7-10μm and 25-30 x 16μm in older colonies.

Alternaria colombiana is unique within the current set of isolates from citrus lesions. Its ex-type isolate, EGS 45-017, originated in Colombia from an old fruit lesion of Minneola tangelo. See Appendix for source information.

233. **Alternaria perangusta** Simmons, sp. nov. Fig. 162

Etym.: L., *per-* + *angusta*, very narrow (of conidia)

Ex cultura in agaro Hay descripta. Coloniae debiliter concentrice zonatae. Conidiophora ad 25-100µm. Catenae conidiorum longissima, constatae ex conidiis 10-20 v. plus. Conidia longe angustiellipsoidea, 15-45 x 3-6µm, conidiophoris secundariis 4-10 x 2-3µm, 2-8 transverse, raro 1 longitudinaliter septata, dilute luteobrunnea, laevia vel punctata. Habitatio typi in laesione Citrus paradisi Macfad. x C. reticulata Blanco cv. Minneola, in arbusto citreo, Sihmrat, Turkey. Typus: partes ex cultura EGS 44-160 (Y. Canihos, YC 8, v.1996 isol.) desiccatae et in ***BPI*** *(holotypus) et saltem in* ***IMI*** *(isotypus) conservandae.*

Colonies on all media used (PCA, Hay, V-8) exhibit rings of growth and sporulation poorly (PCA) or negligibly. Surface and aerial growth on all media is arachnoid to closely interwoven, with both conidiophores and conidial chains so narrow and delicate in appearance that distinguishing between the two at 50X is difficult.

On PCA, **primary conidiophores** and aerial conidiogenous branches commonly are ca. 100-200 x 3-4µm, much the same on V-8, and somewhat shorter on Hay.

Conidial chains on all media become very long, with 10-20+ units, many of which are no wider than the primary conidiophore. Branching of the conidiophore and production of secondary conidiophores and chains from individual conidium cells occurs on all media, but appears to occur more commonly on PCA than on the other media.

On **PCA, conidia** are long-narrow-ellipsoid, rarely wide enough to be termed obclavate, 15-40 x 3-7µm; have secondary conidiophores 3-50 x 2-3µm and 3-7 transepta without longisepta (1 longiseptum in 1 cell of 1 conidium in a chain of 10+ conidia is a rarity); are dilute yellow-tan and usually strongly punctate overall.

On **Hay**, which is used in typification of the species, **conidia** are long-narrow-ellipsoid, 15-45 x 3-6µm; have secondary conidiophores 4-10+ x 2-3µm and 2-8 transepta without longisepta (or rarely 1 longiseptum in 1 cell); are dilute yellow-tan and almost smooth to usually punctate overall.

On **V-8, conidia** are long-narrow-ellipsoid, 15-40 x 3-6µm; have secondary conidiophores 3-15 x 2-3µm or very often

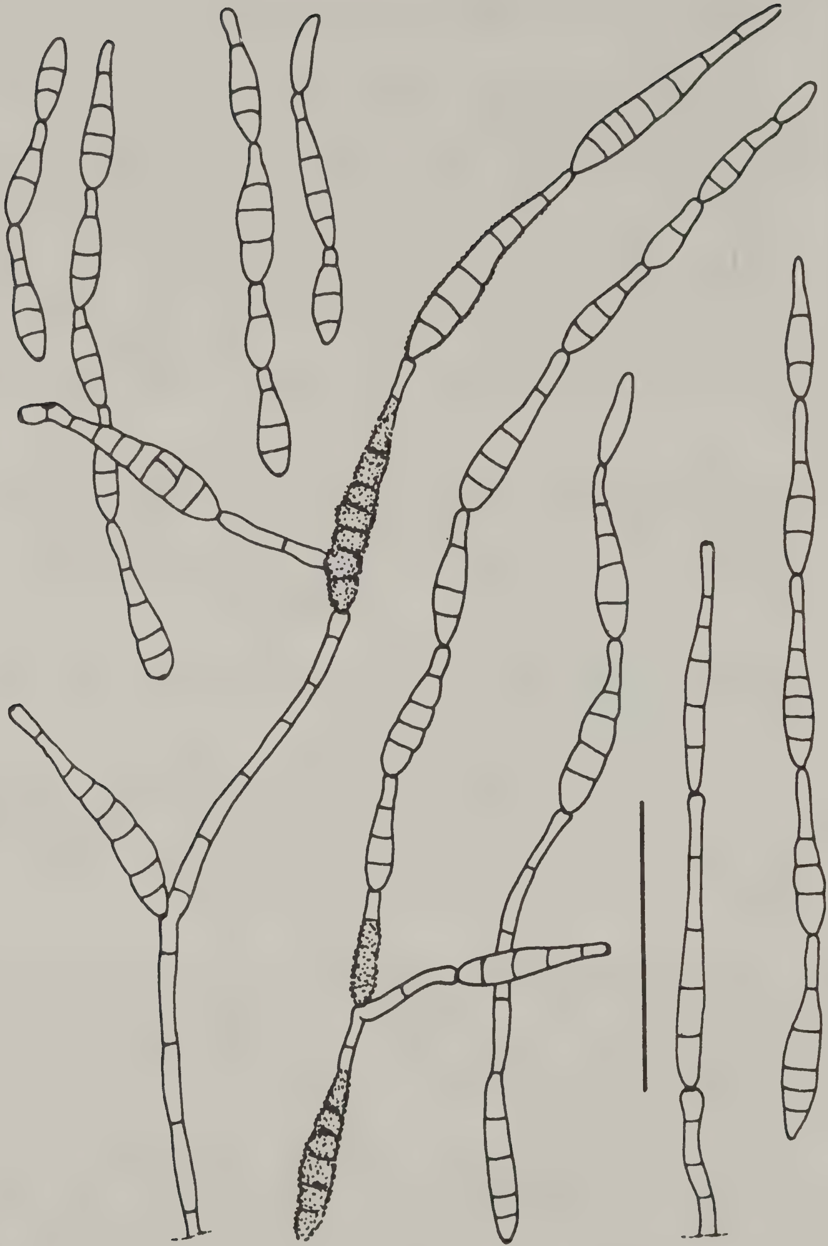

Fig. 162. *Alternaria perangusta*: conidia and conidiophores ex type isolate EGS 44-160. At left, branching chain at 10d. on PCA; at right, simple chains at 5d. on PCA. Bar = 50µm.

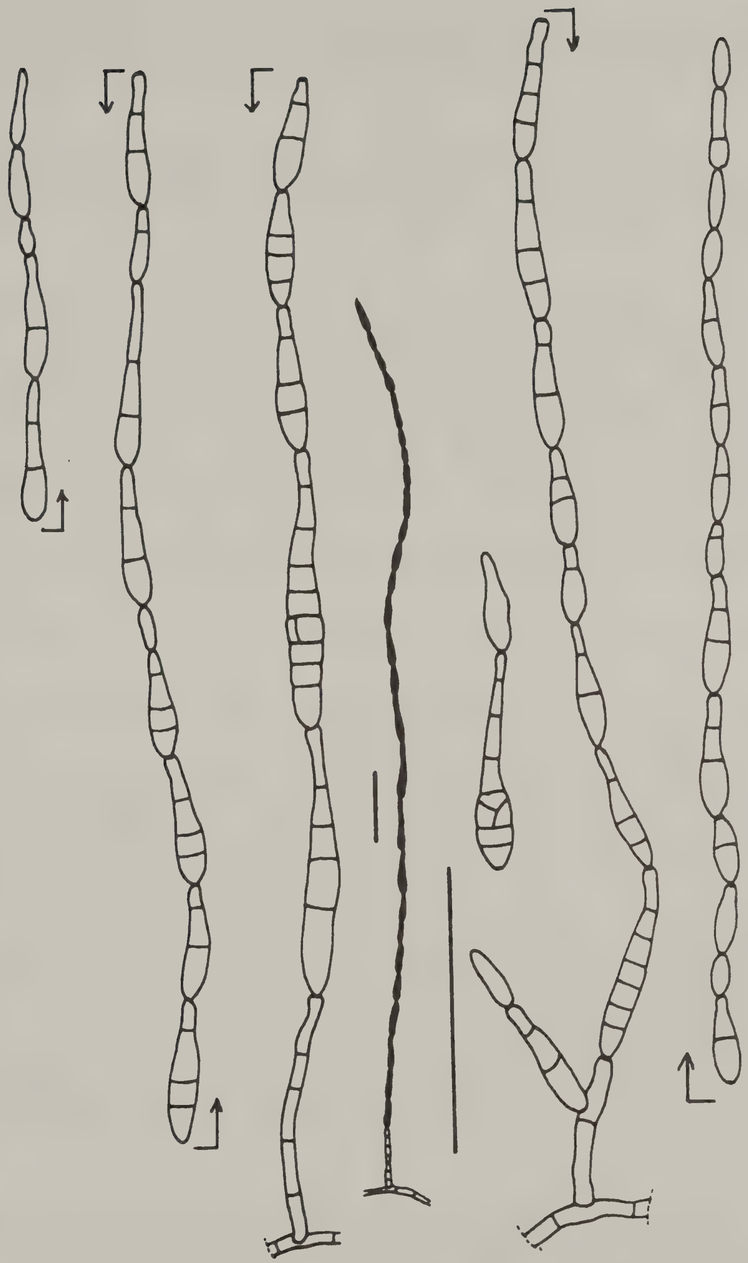

Fig. 162 contd. *Alternaria perangusta*: conidia, conidiophores, habit ex type isolate EGS 44-160. At left, 10d. on Hay; at right, 10d. on V-8. Bars = 50μm.

only a pore in the apex of the preceding conidium, and 1-5 transepta and usually no longisepta (rarely 1 in 1 cell of a conidium); are dilute to medium yellow-tan and often appear quite smooth as well as punctate overall.

Alternaria perangusta is represented by a single isolate, EGS 44-160, which originated in Turkey in 1996 from Minneola tangelo. See Appendix for original source information.

Alternaria perangusta produces large numbers of small conidia without longitudinal septa, as does *Alternaria incomplexa* Simmons (1996, *Alt.T&V* 146). The two species are not confusing in culture or under microscopic comparison, in that the sporulation habit of *A. incomplexa* on PCA and Hay is a small cluster of multiple branches and, in contrast to equivalent elements of *A. perangusta*, the conidiophores commonly are somewhat spirally twisted and multigeniculate with several closely spaced conidiogenous sites, and the conidiophore and conidia are relatively thick-walled, darker, and have well-pigmented septa and apical hila.

234. **Alternaria interrupta** Simmons, sp. nov. Fig. 163

Etym.: L., *interrupta*, abruptly discontinuous (of conidium size and shape in a single chain).

*Ex cultura in agaro V-8 descripta. Coloniae fortiter concentrice zonatae. Conidiophora ca. 40 x 4µm (in luce), 100+ x 4µm (in statu obscuro). Catenae conidiorum interruptae constatae primo ex conidiis 2-3 relative amplis, tum abrupte conidiis additiciis 10-12 perparvulis. Conidia ampla 25-30 x 5-8, conidiophoris secundariis 2-8 x 2-3µm, 3-8 transverse, 1-2 longitudinaliter septata; conidia parva 5-10 x 3-5µm, conidiophoris secundariis parvissimis vel obsoletis, transverse 0-1 septata, longitudinaliter eseptata, dilute sordide brunnea, laevia. Habitatio typi in laesione Citrus paradisi Macfad. x C. reticulata Blanco cv. Minneola, in arbusto citreo, Bet Lyd 29, Israel. Typus: partes ex cultura EGS 45-011 (Z. Solel 11; accept. xi.1996) desiccatae et in **BPI** (holotypus) et saltem in **IMI** (isotypus) conservandae.*

Colonies exhibit concentric rings of growth and sporulation, moderately on PCA and Hay, strongly on V-8.

On **PCA**, **primary conidiophores**, as erect branches of radiating surface hyphae (light exposed), are abundant but distinct and uncrowded; minor numbers of taller primary conidiophores (dark phase) have a few lateral and abundant terminal branches. Surface conidiophores and aerial branches tend to be short, up to ca. 40 x 4μm, but also may lengthen to ca. 100 x 4μm.

On **PCA, conidial chains** usually are simple, with 10-15 conidia, of which the initial 3-4 are comparatively large and the remainder abruptly smaller or at least rapidly decreasing in size. Initial conidia of a long chain are narrow-ellipsoid or narrow-obclavate, 35-40 x 7-8μm, with secondary conidiophores 3-8 x 2-3μm; have 7-8 transepta and 1 longiseptum in 0-3 of the transverse segments. Conidia in the upper part of the chain characteristically are only 7-12 x 4-6μm, with a very short or essentially obsolete secondary conidiophore; have 0-3 transepta and rarely 1 longiseptum in 1-2 conidia of a chain. Terminal conidia in chains whose spores decrease in size gradually (rather than abruptly) usually are ovoid. Conidia are medium dull tan and usually punctate overall.

On **Hay, primary conidiophores** are more abundant than on PCA; they remain distinct or become tangled as their chains elongate; they, as well as aerial branches of taller (dark phase) growth, mostly are short, ca. 40 x 4μm.

On **Hay, conidial chains** are very similar to those on PCA, with the initial 3-5 conidia relatively large, ca. 25-40 x 5-10μm, and the terminal ones usually abruptly or sometimes gradually much smaller, 5-10 x 3-6μm. The larger conidia are narrow-ellipsoid or ovoid; have secondary conidiophores ca. 2-4 x 2-3μm and 3-9 transepta with 1 longiseptum in 1-2 cells of a few conidia; are dull pale tan and smooth to distinctly punctulate.

On **V-8**, which is used in typification of the species, **primary conidiophores** (light exposed) occur in dense stands with overlying masses of dark conidial chains; dark-phase zones of taller conidiophores and their branches have a cottony texture due to the intertwined subterminal branches and their short conidial chains. Primary conidiophores tend to be short, ca. 40 x 4μm, but they and branches in the taller growth zones also may be 100+ x 4μm.

On **V-8, conidial chains** are much like those on PCA and Hay, in that large numbers produce an initial 2-3 relatively large conidia, 25-30 x 5-8μm, then abruptly change to produce 10-12 very small units ca. 5-10 x 3-5μm distal to the large conidia. Chains in which conidium size decreases gradually, from ca. 35-40 x 7-8μm to ca. 10 x 4μm, also are abundant. The large conidia have 3-8 transepta and 1(-2) longisepta in

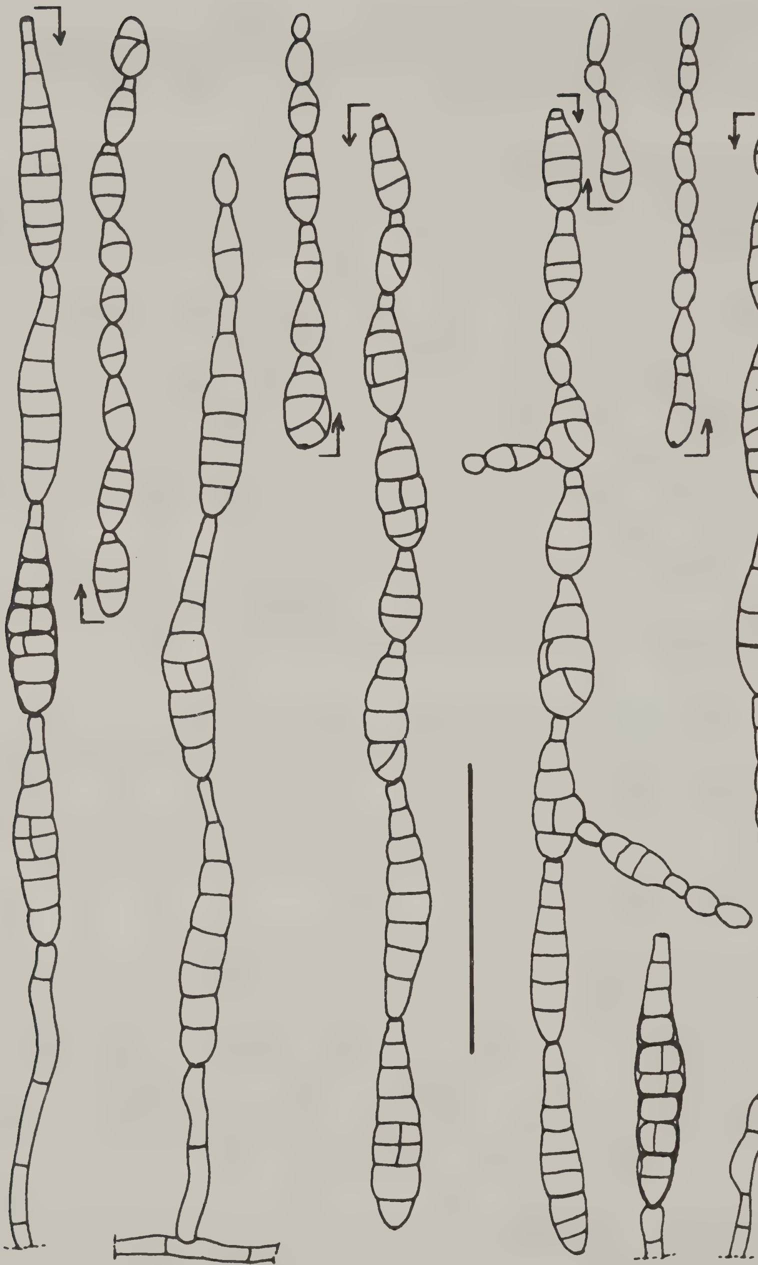

Fig. 163. *Alternaria interrupta*: conidia and conidiophores ex type isolate EGS 45-011. At left, 10d. on PCA; at right, 10d. on Hay. Bar = 50μm.

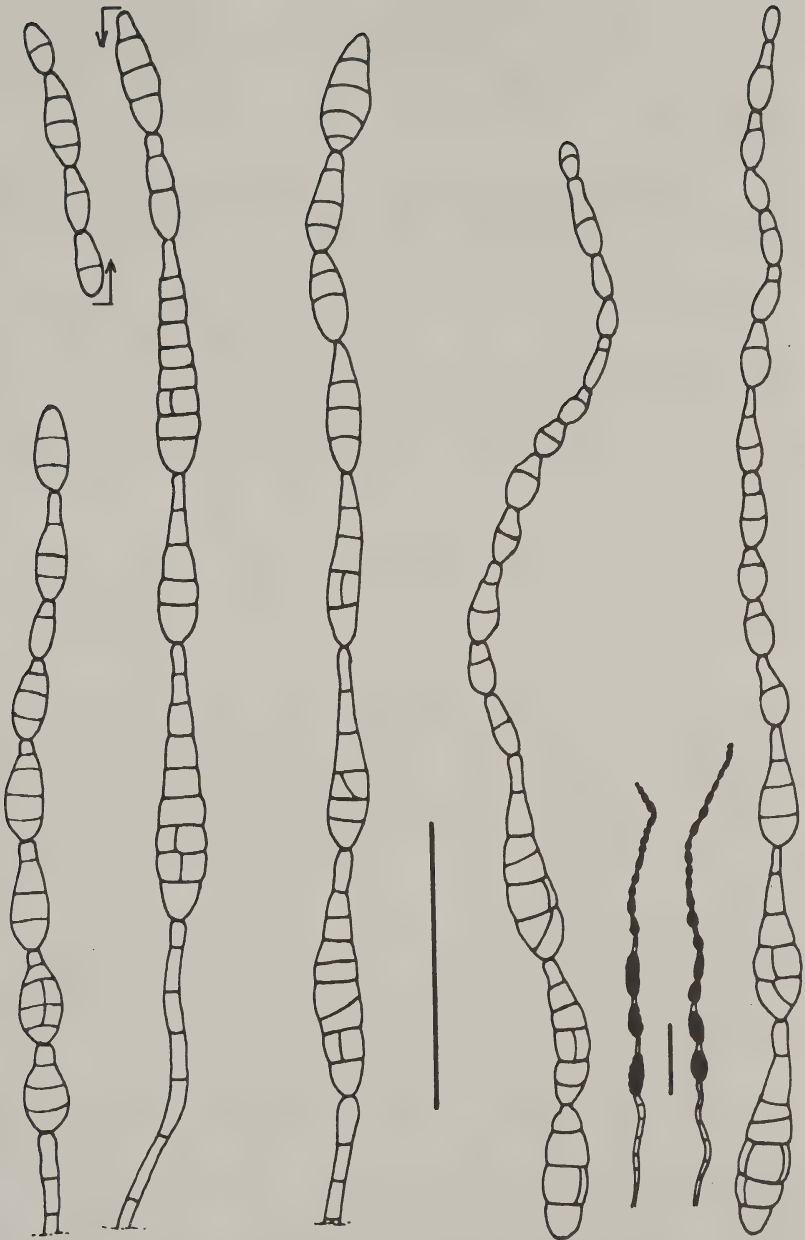

Fig. 163 contd. *Alternaria interrupta*: conidia, conidiophores, habit ex type isolate EGS 45-011. At left, gradual change in conidium size, 5-10d. on V-8; at right, diagnostic abrupt change in conidium size, 5-10d. on V-8. Bars = 50μm.

1-2 transverse segments and secondary conidiophores ca. 2-8 x 2-3µm.; smallest conidia have 0-1 transeptum, no longisepta, and a very small or practically obsolescent secondary conidiophore (an apical pore); and are a medium dull tan and mostly smooth.

Alternaria interrupta is represented by a single isolate, EGS 45-011, which originated in Israel from Minneola tangelo. See Appendix for original source information.

As with many species of *Alternaria* with a variety of conidium sizes and shapes in a controlled population, many individual conidia of *A. interrupta* are morphologically indistinguishable from individuals of other species. However, the interrupted sporulation pattern, initially of large spores then abruptly very small ones, which is visible even at 50X, is characteristic and diagnostic for this species in the context of cultures of relatively small-spored *Alternaria*.

235. **Alternaria dumosa** Simmons, sp. nov. Fig. 164

Etym.: L., *dumosa*, with bushy habit (of sporulation)

Ex cultura in agaro PCA descripta. Coloniae concentrice zonatae. Conidiophora primigena ca. 60-200 x 4-5µm. Ordinatio sporulationis constatae ex conidiophoris secundariis brevibus et catenis conidiorum ramosissimis; catenae conidiorum singulae constatae ex conidiis 4-10; capitulum sporulationis constate ex conidiis 25-200 plus minusve. Conidia ovoidea, 10-20 x 6-10µm, conidiophoris secundariis 3-6 x 3µm vel 25+ x 3µm, 1-3 tranverse, raro 1(-2) longitudinaliter septata, modice aureobrunnea, punctulata vel fortiter ubique punctata. Habitatio typi in laesione Citrus paradisi Macfad. x C. reticulata Blanco cv. Minneola, in arbusto citreo, Mayan Zvi 14, Israel. Typus: partes ex cultura EGS 45-007 (Z. Solel 7; accept. xi.1996) desiccatae et in BPI (holotypus) et saltem in IMI (isotypus) conservandae.

Colonies exhibit concentric zonation in growth and sporulation on all media. Light-exposed areas bear a close stand of bushy clumps of relatively short primary conidiophores with a few short geniculate branches and, at each bend, a short branching chain of conidia. Dark-phase areas produce taller sporulation structures that usually are composed of erect or somewhat reclining bundles of hyphae; these aerial structures are pale but bear pigmented, short conidiogenous branches scattered along their length and at their tips. The taller sporulating elements occur in fairly well-defined rings but also arise scattered among the shorter bushy conidiogenous clumps. Solitary, unbranched conidiophores and conidial chains are uncommon.

On **PCA**, which is used in typification of the species, **primary conidiophores** are ca. 60-200 x 4-5µm with 2-3 geniculate conidiogenous sites. Most of the conidial population is derived from branching secondary conidiophores that develop laterally, basally, and apically from individual cells of the spore body. Individual **conidial chains** have ca. 5-10 units, but the aggregate of spores in the bushy head on one primary conidiophore varies from 25-50 units to commonly 4-5 times that number.

Conidiophore branches on the taller aerial structures are ca. 40-100+ x 4-5µm, and usually have only 1-3 conidiogenous sites, which produce short chains or small clumps of 4-12 conidia.

On **PCA**, **conidia** are mostly ovoid, 10-20 x 6-10µm with secondary conidiophores 3-6 x 3µm or 25+µm long when geniculate and conidiogenous at each bend; have 1-3 transepta and 1 longiseptum in only 1(-2) cells of a few conidia; are medium gold-brown and punctulate to strongly punctate overall.

On **Hay**, **primary conidiophores** usually are shorter and more crowded than on PCA, and produce clumps of fewer conidia. Somewhat open clumps of branching conidiophores and chains commonly have ca. 20-50 conidia. **Conidia** are ovoid or ellipsoid with short or obsolescent secondary conidiophores; have 1-4 transepta and 1 longiseptum in 1(-2) cells of very few conidia in a head, and are dull tan and smooth.

On **V-8**, **primary conidiophores** and sporulating branches of the taller bundles of hyphae are ca. 50-100 x 4-5µm; they may be simple or commonly have 1-4 geniculate conidiogenous extensions. Individual conidial branches are short, with 4-8 units, but branching of the primary conidiophore as well as internal branching of chains by means of terminal, basal, and lateral secondary conidiophores (some of them 1-3 geniculate and sporogenous themselves) generates bushy heads of conidia with 100-200 countable units or usually much higher numbers.

On **V-8**, **conidia** are ovoid or ellipsoid with secondary conidiophores ca. 3-7 x 3µm; have 1-4 transepta and 1 longitudinal or oblique septum in 1(-2) cells of very few conidia in a head; are medium dull brown with strongly pigmented septa, and are mostly smooth, occasionally punctulate to punctate overall.

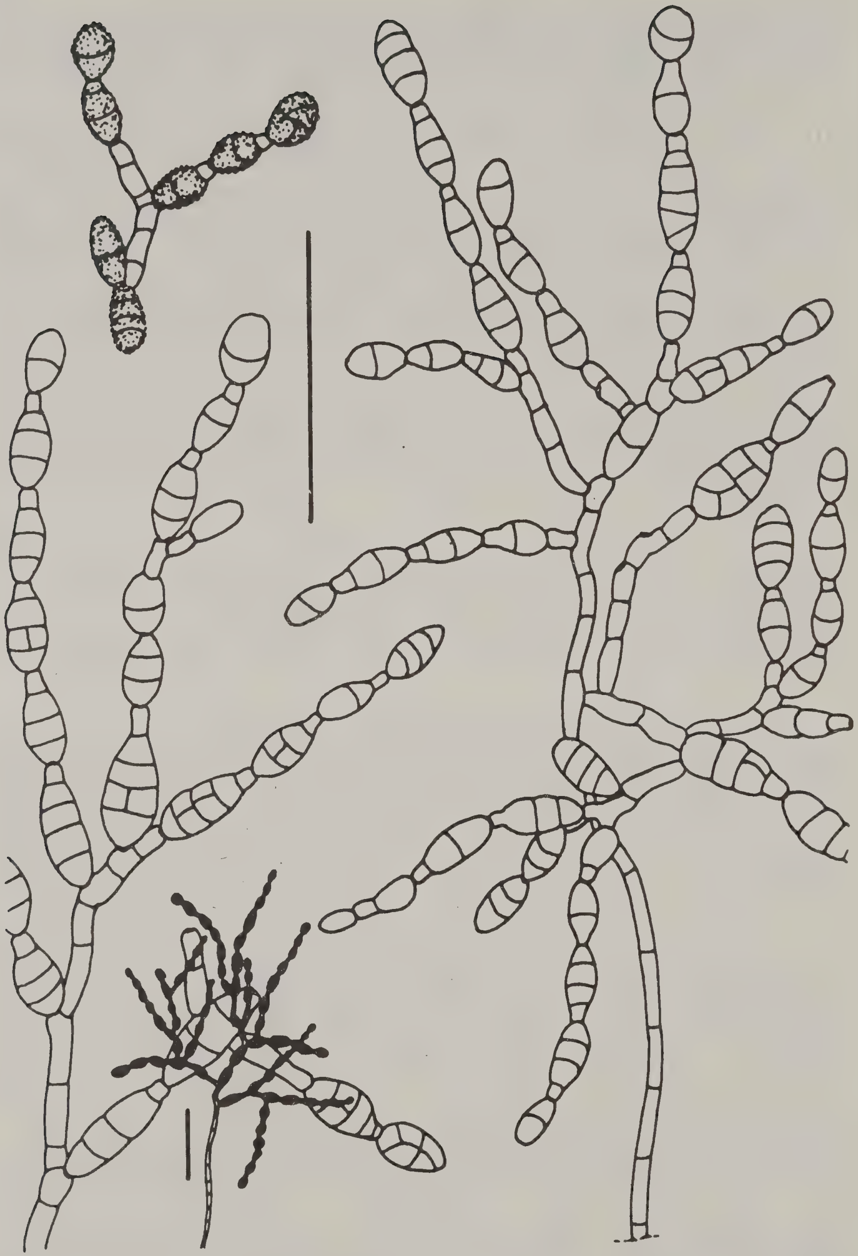

Fig. 164. *Alternaria dumosa*: conidia, conidiophores, habit ex type isolate EGS 45-007. At left, ca. 1/4 of a small sporulation head, and habit of a small head, 10d. on PCA; at right, ca. 1/2 of a small head, 10d. on Hay. Bars = 50μm.

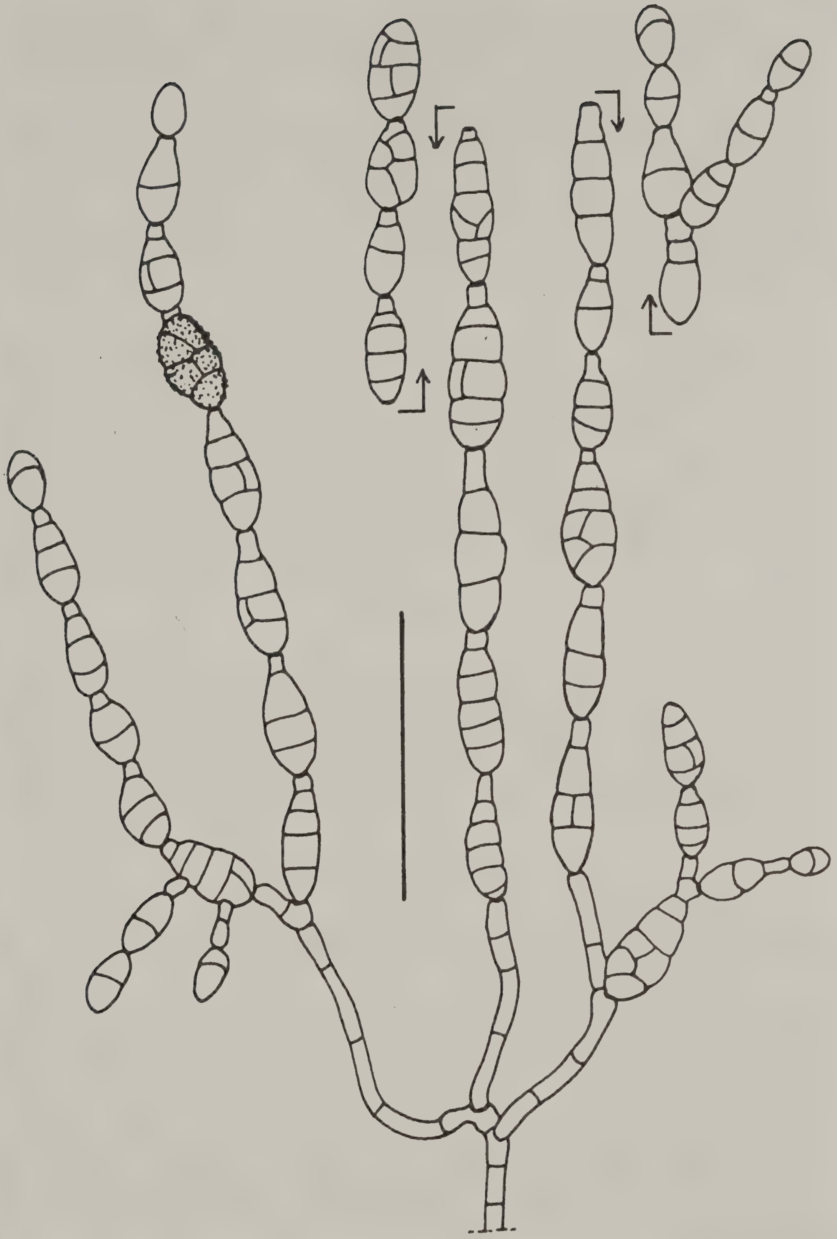

Fig. 164 contd. *Alternaria dumosa*: conidia and conidiophores ex type isolate EGS 45-007; 10d. on V-8 (primary conidiophore branches splayed uncharacteristically for purpose of illustration). Bar = 50μm.

Alternaria dumosa is represented here by a single isolate, EGS 45-007, which originated in Israel from Minneola tangelo. See Appendix for original source information.

The species is unusual in this study set of isolates from citrus in that it is the only one that exhibits a sporulation pattern at all similar to that of typical *A. alternata*. *A. dumosa* has a more condensed sporulation habit and considerably smaller conidia in general than does *A. alternata*. The two are somewhat similar only in a sporulation-pattern sense.

However, *A. dumosa* has a closer resemblance to *A. hesperidiorum* (Pantanelli) Simmons (1990), in that both produce dense heads of short, branching, multigeniculate secondary conidiophores bearing tightly branching chains of small conidia. *A. hesperidiorum* is known at present only from dried voucher specimens (see Simmons 1990; *Alt.T&V* 30); conidia of the species typically are quite smooth, translucent, and to a noticeable extent larger than those of *A. dumosa*. The regional sources of *A. hesperidiorum* (Australia, U.S.A., Cyprus, and New Zealand) suggest that the species probably is common in citrus groves.

Another isolate from navel orange (EGS 39-090), discussed as "*Alternaria alternata* group; species 1" (Simmons 1990), is very similar to *A. dumosa* in sporulation pattern and conidium morphology, including wall ornamentation. Comparison of living isolates of *A. dumosa*, *A. hesperidiorum* and other isolates with tight, bushy sporulation habit is needed.

Discussion and Conclusions

All but one of the named species discussed here as isolates from lesions of rough lemon and tangelo have a sporulation habit of simple conidial chains, short or long, with only moderate or minor amounts of branching, or none, within the chain. Consideration of this non-bushy habit removes them from any possibility of identification with *Alternaria alternata*, which has a sporulation habit of a primary conidiophore supporting a three-dimensional branching system of short secondary conidiophores and conidial chains. The one exception discussed, *A. dumosa*, has a branching sporulation habit similar to that of *A. alternata*; the two species are readily differentiated on the basis of conidium characters.

The 10 named species represent just over half of the isolates actually examined in this study. Among the remainder are the usual quota of culturally unstable isolates found in any large study set. In addition, there are numerous stable isolates that, to the practiced eye, can be distinguished morphologically from others in the set. As remarked earlier, it would be an endless (not quite), thankless (almost certainly) task to describe and illustrate this diversity, although such morphologically definable diversity does in fact exist within this material.

Assuming that most of the isolates I have examined are indeed citrus-disease related, the taxonomic evidence plus earlier observations (Whiteside 1978) and more recent host-specific toxin work (Kohmoto et al. 1991) suggests that *A. limoniasperae* is the specific and unique incitant of *Alternaria* leafspot of rough lemon in Florida. As early as 1929, Doidge established experimentally that *Alternaria* isolates from a serious leafspot disease in rough lemon rootstock nurseries in South Africa were unique in this pathogenicity; isolates from miscellaneous other citrus cultivars were not pathogenic to rough lemon. I have seen no *Alternaria* isolates from South Africa rough lemon, and thus have no opinion on the identity of her material.

With the remarkable, species-identifiable diversity of taxa (named or not) related to brown spot of tangerine and related cultivars, specifically tangelo, the inescapable conclusion is that this widespread disease is a pathology that can be induced by several different species of *Alternaria*. Whether the disease-inducing agent is one common to the several *Alternaria* species, or differs among them, it is certain that Minneola tangelo is susceptible to brown-spot-inducing attacks by a multiplicity of *Alternaria* taxa throughout citrus-growing regions of the world.

The host-specific-toxin literature records that there are at least 13 chemically identified *Alternaria* metabolites related to pathologies of rough lemon and tangerine cultivars (Montemurro and Visconti 1992), with still more being announced (Fujii et al. 1998). Since the literature supporting this list usually does not specify the *Alternaria* isolate that furnished the toxin (e.g., Gardner, Kono and Chandler 1986; Kohmoto et al. 1991 is exceptional in listing strain numbers), it is impossible to verify the identity of the isolates, and it is deduced from the casual undocumented references to "host-selective pathotypes of *A. citri*," that they did not do so.

The relationship between a readily identifiable *A. limoniasperae* and the literature on its pathology and toxicity in "*Alternaria* leaf spot of rough lemon" in Florida is established here.

It is my opinion, which is not at all humble in the face of the voluminous biochemical literature on the subject, that the multiplicity of host-specific toxins reported for the "tangerine pathotype" of "*A. alternata*" and/or "*A. citri*" reflects a degree of multiplicity of distinguishable *Alternaria* species that can induce "*Alternaria* brown spot of mandarin"; and further, that a controlled comparison of the isolates used in toxigenicity studies would support this opinion.

Certainly no isolate examined in the present study is identifiable morphologically with typical *A. alternata* or *A. citri*. The proof lies in the production of cultures under controlled, repeatable conditions, and examination of their sporulation elements with a microscope. These techniques are recommended to anyone who considers maintaining a position that all of these citrus pathogens (and other HST-producers) are morphologically indistinguishable from *A. alternata*, and that they should carry that name.

The one-species or "collective species" concept, which by now has a basis no stronger than interlaboratory gossip, offers no insight to the overall biology and related literature of each of the individual species submerged in this anti-classification practice. The practice makes no contribution to progressive pathology or mycotaxonomy; it should have been abandoned long ago.

Earlier study of *Alternaria gaisen* Nagano (Simmons 1993, Simmons and Roberts 1993) explored the specific identity of this HST-producing incitant of the black-spot disease of Japanese pear. The present study segregates ten distinctive *Alternaria* taxa, at least two of which (*A. limoniasperae* and *A. toxicogenica*) already are related to two major diseases of citrus through previously published studies (cited above) of their toxicity and pathogenicity by specialists in these fields. A study of some other morphologically distinct species, currently misidentified as *Alternaria alternata* in pertinent literature on HST-producing pathotypes, is presented next in *Alt.T&V* 236-243 (Simmons, 1999).

Acknowledgements

It has been an enlightening experience to be able to study, all at the same time and in detail, this set of 135 isolates from citrus lesions from five different citrus-growing regions of the world. I am grateful to the individuals who contributed fresh cultures to the program (institutional addresses listed below in Appendix): Yeter Canihos (Turkey), Berta Lucia Castro C. (Colombia), Zvi Solel

(Israel), and S. H. Swart (South Africa), in addition to T. Peever, L. W. Timmer and J. O. Whiteside (all three now or formerly associated with the University of Florida Citrus Research and Education Center, Lake Alfred, Florida).

After initial morphotaxonomic opinions were generated, I was given the opportunity to view results of RAPD-analysis data generated in the Lake Alfred laboratories on most of the isolates I have studied. This information, generously shared by Y. Canihos, T. Peever, and L. W. Timmer, is an exciting development in its relation to systematic classification of *Alternaria* species related to citrus pathologies. The molecular information and its interpretation are expected to be published by the laboratory team that produced them.

How does one thank reviewers who are professionally supportive on a long-term basis, and good friends, but who will not let a garbled sentence pass, or an illogical conclusion, much less a misplaced comma, or an omitted reference? Quite simply, by acknowledging their contribution fully and in public.

Mary E. Palm initiated the working relationship with L. W. Timmer and T. Peever that led to this examination of isolates from citrus, and has supported the development of the study in numerous ways.

Both Mary Palm and Rodney Roberts have contributed many hours of review, telephone, and e-mail time to ensure appropriate presentation of the results. Acknowledgement of their contributions here is only a summary of my debt.

Wabash College, Crawfordsville, Indiana, continues to support my work through an appointment as Research Associate in the Biology Department faculty, which yields valued access to library and electronic communication facilities.

APPENDIX: Isolate Sources and Terms

BLC	Berta Lucia Castro C., National Center of Coffee Research (CENICAFÉ), Chinchiná, Colombia
CENICAFÉ	Centro Nac. Investigaciones Café "Pedro Uribe Mejia," Chinchiná, Caldas, Colombia
CREC	University of Florida, Citrus Research and Education Center, Lake Alfred, Florida
Dancy	Dancy tangerine, *Citrus reticulata* Blanco cv. Dancy
Minneola	Minneola tangelo, *Citrus paradisi* Macfad. x *Citrus reticulata* Blanco (grapefruit x tangerine = tangelo) cv. Minneola

rough lemon	*Citrus jambhiri* Lush.
SS	S. H. Swart, Letaba Estates, Tzaneen, South Africa
Whiteside	J. O. Whiteside, (formerly at) University of Florida, Citrus Research and Education Center, Lake Alfred, Florida
YC	Yeter Canihos, Department of Plant Protection, University of Gukurova, Adana, Turkey
ZS	Zvi Solel, Agricultural Research Organization, The Volcani Center, Institute of Plant Protection, Bet-Dagan, Israel

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39-181	Whiteside PR320, isol. June 6, 1975, from Dancy tangerine leaf with brown-spot lesions, Wauchula, Florida, "pathogenicity recently verified"; = ATCC 38963 = TU AC320.
39-185	Whiteside PR325, isol. June 20, 1975, from rough lemon leaf with brown-spot lesions, Avon Park, Florida, "pathogenicity recently verified."
39-187	Whiteside PR702, isol. Aug. 23, 1990, from rough lemon leaf with brown-spot lesions, Lake Alfred, Florida, "pathogenicity recently verified.
41-168	CENICAFÉ "B single chain," ex leaf Minneola tangelo, Chinchiná, Colombia; recd. 4 Jan. 1994.
41-169	CENICAFÉ "B single chain," isol. ex leaf of Minneola tangelo, Chinchiná, Colombia; recd. 4 Jan. 1994.
41-170	CENICAFÉ "B-1*," isol. ex leaf of Minneola tangelo, Chinchiná, Colombia; recd. 4 Jan. 1994.
41-175	CENICAFÉ "D stem," isol. ex stem of Minneola tangelo, Chinchiná, Colombia; recd. 4 Jan. 1994.
41-177	CENICAFÉ "D-1 leaf," isol. ex leaf of Minneola tangelo, Chinchina, Colombia; recd. 4 Jan. 1994.
44-159	YC 3, isol. May 1996, ex Minneola tangelo, Abdioglu, Adana Reg., Turkey, "pathogenic to Minneola tangelo and Dancy."
44-160	YC 8, isol. May 1996, ex Minneola tangelo, Sihmwrat, Adana Reg., Turkey, "path. to Minneola and Dancy."
44-161	YC 11, isol. May 1996, ex Minneola tangelo, Kazikli, Adana Reg., Turkey, "path. to Minneola and Dancy."
44-162	YC 15, isol. May 1996, ex Minneola tangelo, Solakli, Adana Reg., Turkey, "path. to Minneola and Dancy."
44-163	YC 15D, isol. May 1996, ex Minneola tangelo, Solakli, Adana Reg., Turkey, "path. to Minneola and Dancy."

44-164	YC 17, isol. May 1996, ex Minneola tangelo, Yunusoglu, Adana Reg., Turkey, "path. to Minneola and Dancy."
44-166	YC 27, isol. May 1996, ex Minneola tangelo, Kuzucuoglu, Adana Reg., Turkey, "pathogenic to Minneola and Dancy."
44-167	YC 31, isol. May 1996, ex Minneola tangelo, Yassidam, Adana Reg., Turkey, "not path. to Minneola and Dancy."
45-003	ZS 3, isol. ex Minneola tangelo, Hadassim 3, Israel; recd. Nov. 1996.
45-004	ZS 4, isol. ex Minneola tangelo, Hadassim 54, Israel; recd. Nov. 1996.
45-005	ZS 5, isol. ex Minneola tangelo, Hadassim 20, Israel; recd. Nov. 1996,
45-006	ZS 6, isol. ex Minneola tangelo, Hadassim 22, Israel; recd. Nov. 1996.
45-007	ZS 7, isol. ex Minneola tangelo, Mayan Zvi 14, Israel; recd. Nov. 1996.
45-011	ZS 11, isol. ex Minneola tangelo, Bet Lyd 29, Israel; recd. Nov. 1996.
45-014	BLC C-1, isol. ex old corky nailhead lesion of Minneola tangelo fruit, Chinchiná, Colombia; recd. Nov. 1996.
45-015	BLC C-2, isol. ex flower bud of Minneola tangelo, Chinchiná, Colombia; recd. Nov. 1996.
45-016	BLC C-4, isol. ex lesion of Minneola tangelo fruit, Chinchiná, Colombia; recd. Nov. 1996.
45-017	BLC C-7, isol. ex old fruit lesion of Minneola tangelo, Chinchiná, Colombia; recd. Nov. 1996.
45-056	SS 97/6, isol. ex fruit spot of Minneola tangelo, Tzaneen, South Africa; recd. April 1997.
45-057	SS 97/2, isol. ex fruit spot of Minneola tangelo, Tzaneen, South Africa; recd. April 1997.
45-058	SS 97/5, isol. ex fruit spot of Minneola tangelo, Tzaneen, South Africa; recd. April 1997.
45-059	SS 97/4, isol. ex leaf spot of Minneola tangelo, Tzaneen, South Africa; recd. April 1997.
45-080	CREC BC2-RLR-01s, isol. ex leaf lesion of rough lemon, location "BC," Florida; recd. July 1997.
45-081	CREC BC2-RLR-06s, (data as 45-080).
45-082	CREC BC2-RLR-08s, (data as 45-080).
45-083	CREC BC2-RLR-13s, (data as 45-080).
45-084	CREC BC2-RLR-17s, (data as 45-080).
45-085	CREC BC2-RLR-18s, (data as 45-080).
45-086	CREC BC2-RLR-25s, (data as 45-080).
45-087	CREC BC2-RLR-26s, (data as 45-080).
45-088	CREC BC2-RLR-28s, (data as 45-080).
45-089	CREC BC2-RLR-31s, (data as 45-080).
45-090	CREC BC2-RLR-32s, (data as 45-080).

45-091	CREC BC2-RLR-33s, (data as 45-080).
45-092	CREC BC2-RLR-35s, (data as 45-080).
45-094	CREC BC2-RLR-38s, (data as 45-080).
45-095	CREC HC2-RLR-01s, ex leaf lesion of rough lemon, location "HC," Florida; recd. July 1997.
45-097	CREC HC2-RLR-06s, (data as 45-095).
45-098	CREC HC2-RLR-07s, (data as 45-095).
45-099	CREC HC2-RLR-08s, (data as 45-095).
45-100	CREC HC2-RLR-11s, (data as 45-095).
45-101	CREC HC2-RLR-12s, (data as 45-095).
45-103	CREC HC2-RLR-17s, (data as 45-095).
45-104	CREC HC2-RLR-18s, (data as 45-095).
45-105	CREC HC2-RLR-19s, (data as 45-095).
45-106	CREC HC2-RLR-21s, (data as 45-095).
45-107	CREC HC2-RLR-22s, (data as 45-095).
45-114	CREC EV-MIL-2s, isol. ex Minneola tangelo, Lake Alfred, Florida; recd. August 1997.
45-115	CREC EV-MIL-8s, (data as 45-114).
45-116	CREC EV-MIL-9s, (data as 45-114).
45-120	CREC EV-MIL-28s, (data as 45-114).
45-121	CREC IMK-MIL-6s, isol. ex Minneola tangelo, Immokalee, Florida; recd. Aug. 1997.
45-122	CREC IMK-MIL-10s, (data as 45-121).
45-125	CREC IMK-MIL-38s, (data as 45-121).
46-033	CREC BC2-RLR-19s, isol. ex leaf lesion of rough lemon, location "BC," Florida; recd. Oct. 1997.
46-036	CREC BC2-RLR-34s, (data as 46-033).
46-077	CREC HC2-RLR-15s, isol. ex leaf lesion of rough lemon, location "HC," Florida; recd. Jan. 1998.
46-109	CREC HC2-RLR-20s, (data as 45-077).
46-136	CREC SH-MIL-1s, isol. ex leafspot of Minneola tangelo, location "Shinn Grove," Florida.
46-139	CREC SH-MIL-4s, (data as 46-136).
46-140	CREC SH-MIL-8s, (data as 46-136).
46-141	CREC SH-MIL-15s, (data as 46-136).
46-142	CREC SH-MIL-18s, (data as 46-136).
46-143	CREC SH-MIL-19s, (data as 46-136).
46-147	CREC SH-MIL-32s, (data as 46-136).
46-148	CREC SH-MIL-36s, (data as 46-136).
46-149	CREC SH-MIL-37s, (data as 46-136).
46-150	CREC SH-MIL-38s, (data as 46-136).
46-159	Whiteside PR309, isol. leaf of rough lemon, Florida; = ATCC 38962.

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ALTERNARIA THEMES AND VARIATIONS (236-243)
HOST-SPECIFIC TOXIN PRODUCERS

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ABSTRACT

Isolates of six of the major HST-producing species of *Alternaria* are compared in axenic culture under controlled conditions. None of these can be identified morphologically with typical *A. alternata*. The history of an untenable "*Alternaria alternata* pathotype" treatment in an "*Alternaria alternata* collective species" concept for these HST-producers is confronted. The concept should be abandoned; its origin was based on misinterpretation of derivative data in an early Simmons paper, and its continued practice ignores modern approaches to examination and comparison in *Alternaria* systematics.

Representative isolates are described, illustrated, and compared for *A. alternata*, *A. tenuissima*, *A. gaisen* (causing black spot of Japanese pear), *A. limoniasperae* (brown spot of rough lemon), *A. toxicogenica* (one of several incitants of brown spot of tangerine-related cultivars), *A. longipes* (brown spot of tobacco), *A. mali* (apple blotch), and *A. arborescens* Simmons sp. nov. (stem canker of tomato).

Keywords: *Alternaria*, host-specific toxins, "pathotypes"

XXVII. CLASSIFICATION OF ALTERNARIA SPECIES THAT PRODUCE HOST-SPECIFIC TOXINS

Introduction

Observations and conclusions in the preceding *Alt.T&V* 226-235, classification of citrus pathogens (Simmons 1999), raise questions about the taxonomic validity and the practical usefulness of treating several morphologically identifiable species of *Alternaria* simply and confusingly as "pathotypes" of *Alternaria alternata* (Fr.) Keissler (1912).

The practice exists primarily in literature related to research on host-specific toxin (HST) production, with *Alternaria* taxa as major examples of such capability. HST literature, however, pays only minimal attention to differential characters other than the toxins themselves.

The emphasis in HST research is primarily physiology and biochemistry. Its influence, however, has spilled over into reiterative, undocumented statements that all HST-producing *Alternaria* strains are so similar morphologically that they cannot be distinguished from each other. This position is very far from the truth, but it has infected fungus-toxin research to the extent that nearly all related publications submerge their results under one name, *Alternaria alternata*, without a shred of morphological or other comparative data to support the position. Evidence for the one-name position does not in fact exist.

Identification questions that reach me on the subject commonly are some variant of "Where did all of these *A. alternata* pathotypes come from all of a sudden?" The bottom-line answer, of course, is that they did not come from any place "all of a sudden"; they are distinguishable taxa, each with its own bibliographic history and morphologic identity and, in some cases, with its own information base of morphology and pathology already attached to its published name. It is ironic that misinterpretation or misunderstanding of a Simmons paper (1967) on typification of *Alternaria* led to the still unchanged but readily assailable misuse of *A. alternata* as the catchall name for increasingly diverse HST-producing species.

One purpose here is to illustrate and describe the individual species that much of the HST literature lumps under the single name, *Alternaria alternata*, without supporting diagnostic data. As a prelude to this identification and description of most of the pertinent *Alternaria* species, I propose to present a brief history of the origin and practice of using *A. alternata* as an umbrella name for several distinct HST-producing *Alternaria* species; to assess the morphologic, taxonomic, and nomenclatural errors residing in the practice; and to debunk the pretense that the several morphologically distinct HST-producers constitute a one-name, multi-toxin *A. alternata* conglomerate in any sort of rational *Alternaria* taxonomy.

Historical Background: Fiction and Facts

The HST-related literature, which practices the one-species-name-fits-all approach to most of the HST-producing species of *Alternaria*, incorporates at least four theses in its misuse of *Alternaria* classification and nomenclature—none of them valid or tenable. These misconceptions are explored below, with pertinent literature references.

1. **Fiction**: that the seven HST-producers emphasized in current literature, the so-called "pathotypes" of *Alternaria alternata*, are so similar that they cannot be distinguished from each other by morphological examination (Kusaba and Tsuge 1994, 1995; Nishimura 1980, Nishimura and Kohmoto 1983, Scheffer 1992).

Fact: This idea of indistinguishability, which is unsupported by published data, can be disproved easily by anyone who will grow isolates under specific controlled conditions, and who can use a microscope. In evidence are the descriptive, illustrated treatments of the several species presented below.

The one-species-name practice was introduced by Nishimura and his colleagues and students (1978) in a report of work on the HST-producing pathogen that induces blackspot of Japanese pear. The choice was based solely on their inability to distinguish isolates of *A. gaisen* Nagano (1920; = illegitimate *A. kikuchiana* Tanaka, 1933) from published conidium dimensions of the type specimen of *A. alternata*, as presented by Simmons (1967). Since *A. alternata* was the correct name for the earliest known small-spored species of the genus, they adopted it for all of their material.

The choice of name has not been reviewed in HST practice, even when other HST species with quite different morphological and biological properties entered the field. Thus, progress in definitive *Alternaria* studies based on pure culture comparisons has not been paralleled by progress in HST thinking, which has remained locked in a collective-species mold based on misunderstanding of the 1967 Simmons material. HST work has expanded from the initial study of *A. gaisen* to an undocumented position that the seven or so posited HST-producers cannot be differentiated from each other or from *A. alternata* on morphological grounds. The facts to the contrary are established below.

2. **Fiction**: that measurements of conidia from the type specimen of *Alternaria alternata*, as presented by Simmons (1967), are sufficiently definitive and indistinguishable from those of conidia of the "pathotypes" that the name *A. alternata* should be used for the entire miscellany of HST-producing species (Nishimura 1980, Nishimura et al. 1978, Nishimura and Kohmoto 1983).

Fact: This represents a complete misunderstanding of the relationship between the size of a few dissociated conidia of a type specimen at least 150 years old and the sporulation characteristics of morphologically diverse, active, pure cultures in modern times.

It now is indefensible (and should have been in 1978 and subsequently) to cite Simmons and maintain that his 1967 discussion of the attributes of the type specimen of *A. alternata* is a rational basis for defining the species as it is known through methods of axenic culture, microscopy, and biochemical investigation.

The distinction between typification of a fungus name and subsequent characterization of the species itself often is missed—not only by clients of taxonomic information but by many taxonomists themselves. The conidium population and its configuration in hundreds of *Alternaria* type specimens from field collections (and in the thousands for other genera) frequently represent a unique event, not a repetitive or progressive one, in the development of the specimen—often simply a quick overnight flush of sporulation induced by microclimate changes.

Morphology data of that population, if considered distinctive within the genus, can be published and assigned a name. The name and the original data, which typify the name, are forever linked in some way when we comply with internationally agreed rules of nomenclature (Greuter et al., 1994). However, measurements of sporulation elements in a type description are static—they do not define the limits or the expression of characters observable under growth conditions different than the original. The working description of any species, at any moment, is the accumulation of reproducible observations on its multiple characters under multiple conditions of growth, in nature or in culture. The concept of a species, signified by use of its name, always must include the type characters. But those static type characters, e.g., conidium measurements, do not define the species; and equally importantly, they cannot be used as a unique template for identifying other specimens from field or culture.

Using type conidium measurements as an exclusive identification character, in the manner HST workers have done with *A. alternata*, would lead to chaos in research and applied work if directed, for example, to *Aspergillus*, *Fusarium*, *Penicillium*, *Cladosporium*, *Trichoderma*, and large numbers of other genera. *Alternaria* cannot be the exception.

The original Nees field specimen (EGS 11-050), the type of *A. alternata* dating from the early 1800s, yielded ca. 25 conidia whose description typifies the name, but which go no further in characterizing the species. Study and interpretation of additional specimens have permitted specialists in the genus to interpret the biological nature and measurable characteristics of the species, which inevitably are an expansion of type specimen data, as is true for every organism known to science. The current Simmons concept of *A. alternata* is defined by characteristics of stable isolates as well as the type specimen (Simmons 1992, 1995; Simmons and Roberts 1993); it incorporates the basic (but not "collective species") usages of the other two most cited authors, Neergaard (1945) and M. B. Ellis (1971). See item 3, following.

3. **Fiction**: that "these fungi [HST pathotypes] look similar in conidial morphology, and statistically the measurements of conidial size fall within a category of the descriptions of a collective species *Alternaria alternata* (Fries) Keissler..., as described by Simmons [1967]" (Nishimura and Kohmoto 1983, paraphrasing Nishimura 1980, in turn based on Nishimura et al. 1978); and that this contention supports their *A. alternata* multi-pathotype usage in HST literature.

Fact: Either a factual misstatement was made with reference to Simmons—who has never held or published a "collective species" concept of *A. alternata*—or there was a transfer from some other source, without attribution or citation. But most certainly it did not come from Simmons, either then or at any subsequent time.

The term "collective species," used in the HST literature but never used or implied by Simmons, has no meaning in taxonomic theory or species-differential practice. It seems likely that the term "collective species," whose meaning is never defined or explained in HST usage, was borrowed from Neergaard (1945). I have not found the term in use elsewhere in major *Alternaria* taxonomic literature, although M. B. Ellis, in his manual of dematiaceous hyphomycetes (1971), must have used such an encompassing concept for practical purposes in his presentation of *A. alternata*, as becomes evident when large numbers of his identified specimens are re-examined.

Neergaard, in his rich and useful presentation of *Alternaria* species known to him in Denmark, did not ever define or settle on one sporulation pattern or representative isolate as typical of *A. alternata* (= synonymic *A. tenuis* in Neergaard's writing). Instead, he followed the Wiltshire

(1933) practice of applying a designation "*Alternaria 'tenuis' auct*....to the group of *Alternaria* forms which of recent years has been mentioned in the mycological literature under the name *A. tenuis*" (Neergaard, l.c.,p. 87).

The crucial factor, when taking guidance or quoting from Neergaard's work, lies in his warning (l.c., p. 87) that: "It is the collective species *Alternaria tenuis auct*. that will be dealt with in the following." Throughout his treatment of "*A. tenuis auct.*" he incorporated a range of sporulation types and small-spored morphology that he believed met the various "*A. tenuis*" ideas of several different publishing authors ("*auct.*"). He did not select one "*A. tenuis*" isolate to represent his observations; nor did he ever express an opinion on what might constitute typical *A. alternata*. In his only restricting or defining comment on the subject, he used a term "*A. tenuis auct. sensu stricto*," which referred (l.c., p. 89) to examples "with spores of pronounced variable form, without beak or with a usually very short and rather broad beak." I retain this same defining concept for conidia of *A. alternata* in controlled culture, adding stable characters of branching sporulation pattern and of conidium development, size, and septation (e.g., as in widely available isolate EGS 34-016). Neergaard's only restrictive evaluation of *A. alternata* conidia, which is essentially the same as mine, is abundantly illustrated in his Figures 8 and 9 (l.c., pp. 96-97).

This digression into the source of the term "collective species" emphasizes the original meaning, namely, the variety of "*A. tenuis*" (= *A. alternata*) concepts of several different authors of Neergaard's time; this precludes the misinterpretation that such a multiple concept can be transferred *in toto* to a one-species taxon named *A. alternata*. HST literature has employed this "collective species" concept for 20 years in the manner of a thoughtless cliché, mistakenly attributing its concept to me. As introduced by Neergaard, it has no species-transferable meaning. As used in HST literature, it has the same meaning, i.e., none.

4. **Fiction**: that *A. alternata* is ubiquitous and/or the most commonly encountered of saprophytic *Alternaria* species (Nishimura and Kohmoto 1983, Scheffer 1992, Kusaba and Tsuge 1994). This idea is parroted in one form or another in nearly every HST-related publication cited here, as well as in great numbers of survey lists of isolates from crops, soil, air, etc., whose reliability becomes questionable at the minute they list "*A. alternata*."

Fact: A statement of this sort appears as introductory to many HST publications, without a particle of internal evidence or authoritative reference entered in its support.

Typical *A. alternata* is defined at a minimum as a species with a three-dimensional sporulation pattern of a relatively short primary conidiophore that bears a branching system of secondary conidiophores and chains of small conidia with species-defining characters (Simmons 1990, 1992, 1995, 1999; Simmons and Roberts 1993).

The species, defined thus, is not encountered frequently among many hundreds of small-spored isolates studied in comparative culture from multitudes of sources, common or rare. In a study involving 349 *Alternaria* isolates from Japanese pear and apple lesions, most of them non-pathogenic to Japanese pear (Japan, Korea, Taiwan, U.S.A.), only seven had a sporulation pattern at all similar to that of *A. alternata* (Simmons and Roberts 1993). And in the recently reported examination of 135 *Alternaria* isolates from citrus lesions (Colombia, Israel, Turkey, South Africa, U.S.A.), not a single isolate could be equated with typical *A. alternata* (Simmons 1999).

The species *A. alternata* is not commonly encountered; misidentifications are superabundant. It is easy to run a computer count of the number of times the name appears in the literature; it is quite another matter to verify the correctness of the identifications. An estimate of correct identification, based on attempts to verify hundreds of contributed cultures, approaches zero.

Confronting the Collective Species Concept

The *A. alternata* "collective species" concept, as used in HST literature, was challenged by Simmons and Roberts (1993), when it was demonstrated that one of the first well-studied HST producers, *Alternaria gaisen* Nagano (1920), has no character of sporulation production or conidium morphology that would permit its identification as *A. alternata* in the sense of frequently cited specialists on classification within the genus (M. B. Ellis 1971; Neergaard 1945; Simmons 1990, 1992). *Alternaria gaisen*, sometimes known incorrectly as *A. kikuchiana* Tanaka (1933), usually is treated in the HST literature as "Japanese pear pathotype of *A. alternata*" (Nishimura 1980) or some minor variant of this phrase.

The *A. alternata* one-name, collective-species usage is challenged even more broadly in *Alt.T&V* 226-235 (Simmons 1999). Several morphological segregates within a large set of 135 pathogenic isolates from *Citrus* species are defined there individually as species; all of them lack sporulation resemblance to typical *A. alternata*. For example, *A. limoniasperae* Simmons and *A. toxicogenica* Simmons, newly described in that place, have long figured in the HST literature, respectively as "*Alternaria alternata* rough lemon pathotype" and "... tangerine pathotype" (Nishimura and Nakatsuka 1989), even though no morphological evidence of such an "*A. alternata*" relationship has ever been published.

The "*A. alternata* collective species" conceit has been quoted from paper to paper in the HST literature for two decades. The phrase has no meaning in critical taxonomic analysis. If it means something to HST practitioners, their use of it should be explained. Taxonomic opinions, like opinions in any scientific discipline, require published evidence; nothing pertinent exists in the HST literature.

Species of HST-producing Alternaria

Six of the seven *Alternaria* species reported to produce host-specific toxins are illustrated and described here. Typical *A. alternata* and *A. tenuissima* are added for purposes of contrast. The one HST-related omission is the so-called "strawberry pathotype"; I have seen numerous morphologically diverse isolates from strawberry lesions (Australia, Belgium, Canada, Italy, New Zealand, U.K., U.S.A.) but not examples from HST-related strains involved in "black spot of strawberry" in Japan.

It does not pass unnoted that Yu (1992) refers to his examination of an HST-producing strawberry pathotype as "*A. fragaria*," with his opinion that isolates "displayed enough characteristics in common with standard stock cultures of *A. alternata* [including EGS 35-193]...to be considered as the same species"; and that Bottalico and Logrieco (1998) list "*A. fragariae* Dingley (= *A. alternata* strawberry pathotype)."

No *Alternaria* taxon has ever been described as *A. fragaria(e)*. J. Dingley (1970) reported and described "*Alternaria alternata* (Fr.) Keissler f.sp. *fragariae*" causing leaf lesions of strawberry in New Zealand; her isolates, which I later examined and discussed with her, are species of *Embellisia*, not *Alternaria* (see Simmons 1992, p. 26-27). How confusing the *Alternaria* literature must be to strawberry pathologists, when reported pathogens, with "pathotype" attributions, represent two different hyphomycete genera. Presumably the HST strawberry work is with an *Alternaria*; if so, it has nothing to do with Dingley's "f.sp. *fragariae*." A taxonomic mix-up, therefore an informational mess.

All isolates discussed here were grown and studied under identical conditions: PCA, Hay, V-8 agars in sectors of 90mm Y-plates; 22-24C; ambient, gradually drying atmosphere; cool-white fluorescent light cycled daily 8hr/16hr light/dark. All comparisons are based on PCA cultures, with comments from either Hay or V-8 added when an additional diagnostic clue is relevant. Sporulation patterns and conidia were studied at 50X, 195X and 780X.

The species are presented in the following order, with "pathotype" designations used in Kohmoto and Durbin (1989), plus other referent phrases.

236. *Alternaria alternata* (Fries) Keissler (1912)

237. *Alternaria tenuissima* (Nees : Fries) Wiltshire (1933)

238. *Alternaria gaisen* Nagano (1920)
= *A. kikuchiana* Tanaka (1933)
"*A. alternata* Japanese pear pathotype"
incitant of black spot of Japanese pear

239. *Alternaria limoniasperae* Simmons (1999)
"*A. alternata* rough lemon pathotype"
"*A. citri* rough lemon pathotype"
incitant of brown spot of rough lemon

240. *Alternaria toxicogenica* Simmons (1999)
"*A. alternata* tangerine pathotype"
"*A. citri* tangerine pathotype"
incitant of brown spot of tangerine

241. *Alternaria longipes* (Ellis & Everhart) Mason (1928)
"*A. alternata* tobacco pathotype"
incitant of brown spot of tobacco

242. *Alternaria mali* J. W. Roberts (1914)
"*A. alternata* apple pathotype"
incitant of *Alternaria* blotch of apple

243. *Alternaria arborescens* Simmons sp. nov.
A. alternata f.sp. *lycopersici* (Grogan et al. 1975)
"*A. alternata* tomato pathotype"
incitant of stem canker of tomato

* * * * *

Instead of noting the morphologic difference of *Alternaria alternata* from each HST-related species when it is described, drawings of sporulation patterns and diagnostic mature conidia of all species are summarized in Fig. 173 as part of the final Discussion and Conclusions.

* * * * *

236. ***Alternaria alternata*** (Fr.) Keissler (1912)   Fig. 165
= *Alternaria tenuis* Nees (1816)

Background Information

In the work of the three most cited students of *Alternaria* systematics, Neergaard's major publication on species known to him in Denmark (1945) did not concern itself with typification of *A. alternata* or its correct name (he used synonymic *A. tenuis*). M. B. Ellis (1971) used the typification and nomenclature established in Simmons (1967), where the correct nomenclature, typification, and illustrations of type conidia were presented.

Neergaard's treatment of *A. alternata* was in terms of a group of *Alternaria* forms mentioned in the literature by several other authors under the name *A. tenuis*. He referred to this miscellaneous group of identifications as a "collective species" and made a specific point of stating that his extended discussion of "*A. tenuis*" actually was for this miscellaneous "collective species" sense of many different authors. He did not establish or promote a one-species concept for *A. tenuis* (*A. alternata*) in his work.

Ellis, in his initial manual of dematiaceous hyphomycetes (1971), also practiced a "collective species" approach to the large numbers of specimens he identified as *A. alternata*. He was not a specialist in the systematics of small-spored species of *Alternaria*, nor did he pretend to be. For practical purposes of manual preparation, Ellis, like Neergaard, did not maintain a closely-defined concept of typical *A. alternata*. However, his illustrations of the species (l.c., p. 465) present conidia and a sporulation pattern that are close to what I deduce Neergaard and I both would accept for this type species.

Simmons, in 30 years of a monographic approach to *Alternaria* taxonomy, has maintained a closely defined view of the morphologic entity *A. alternata* as it develops in culture under specific controlled conditions. To do otherwise, as does a "collective species" approach, would only continue the chaos that exists in the systematic treatment and identification of morphologically distinctive small-spored, chain-forming species of *Alternaria*.

By defining *A. alternata* in a sense that incorporates conidia of the type specimen (from field material) as well as isolates with relatively small conidia, short secondary conidiophores, and a habit of branching chains of conidia, it has been possible to select representative strains from among the large numbers of cultures available to me.

It is true, of course, that some set of isolates other than those indicated below could have been chosen as comparison standards for *A. alternata*. However, this still would not have solved the fundamental problem of *Alternaria* systematics, in that there are large numbers of morphologically distinct and culturally stable segregates among small-spored *Alternaria*; if one taxon—any one—is chosen as *A. alternata*, then all the others become no less distinct and diagnosable as named species.

An early decision in HST-related work—i.e., to use the name *A. alternata* for morphologically distinctive *A. gaisen* (Nishimura et al. 1978)—was injudicious, and became increasingly unacceptable as other HST-producing species with well-established names, or with quite different morphologies and unique biological properties, were gathered under the same name.

The excuse/explanation for this curious one-name usage always has been that conidium measurements of HST-producers cannot be distinguished from those of a few conidia of the typical *A. alternata* field specimen (Simmons 1967). This is not an erudite position to hold in this age of axenic culture techniques. No practicing hyphomycete systematist would make such a statement, particularly when there is access to even minimal culture plate and microscope facilities. It is easily demonstrable to any biologist with an enquiring mind on the subject that not one of the major HST-producing species can be identified with *A. alternata*.

Isolate Used for Description and Illustration

A few years ago Simmons selected, from among large numbers of cultures available to him, four isolates that appeared morphologically identical and culturally stable under controlled conditions. These strains were cited as representative *A. alternata* (Simmons 1992, 1995). They are available from several international culture collections (e.g., ATCC, CBS, IFO, IMI). Since that time, one of the four has become unsuitable for comparison work.

The three isolates recommended at present as representative are EGS 34-016, 34-039, and 35-193. Strain 34-016 is

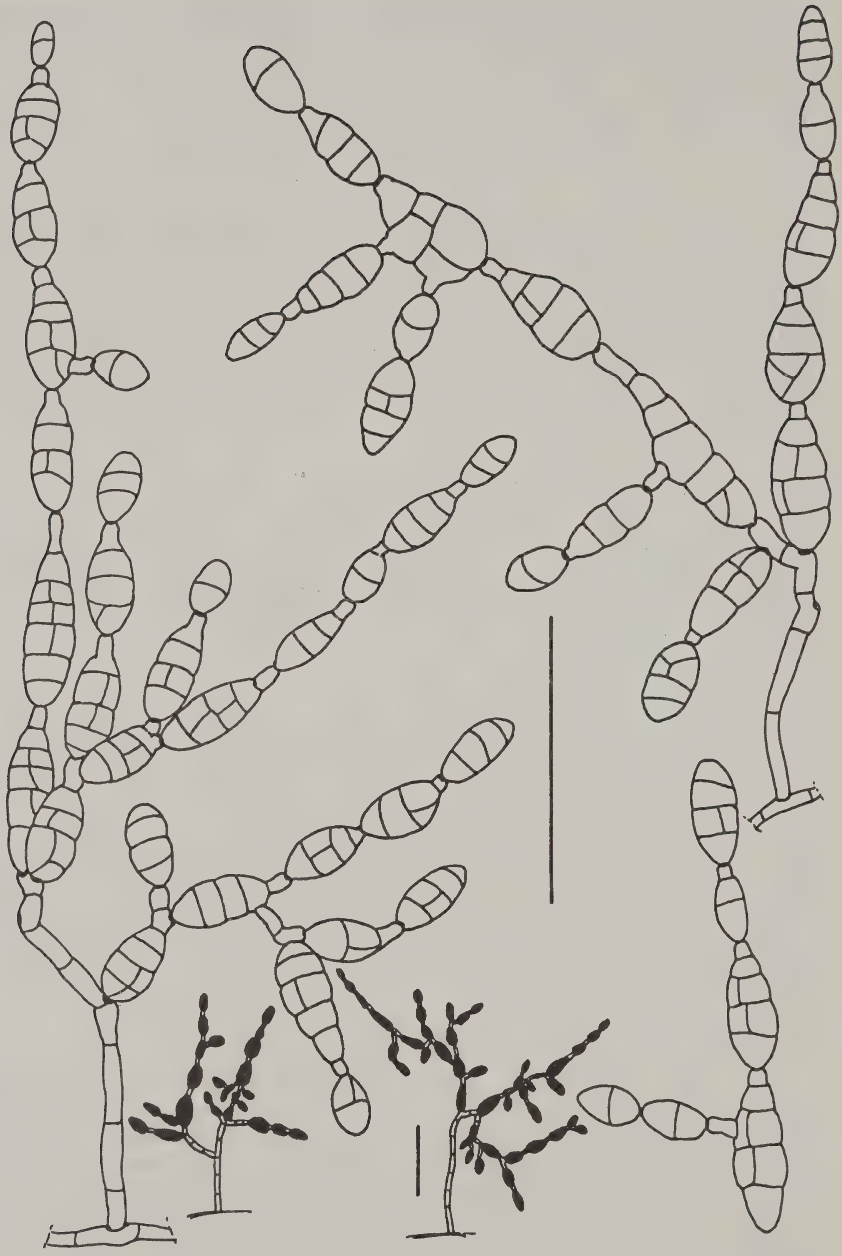

Fig. 165. *Alternaria alternata*: portions of sporulation clumps and habits from PCA (at left) and Hay (at right); EGS 34-016. Bars = 50μm.

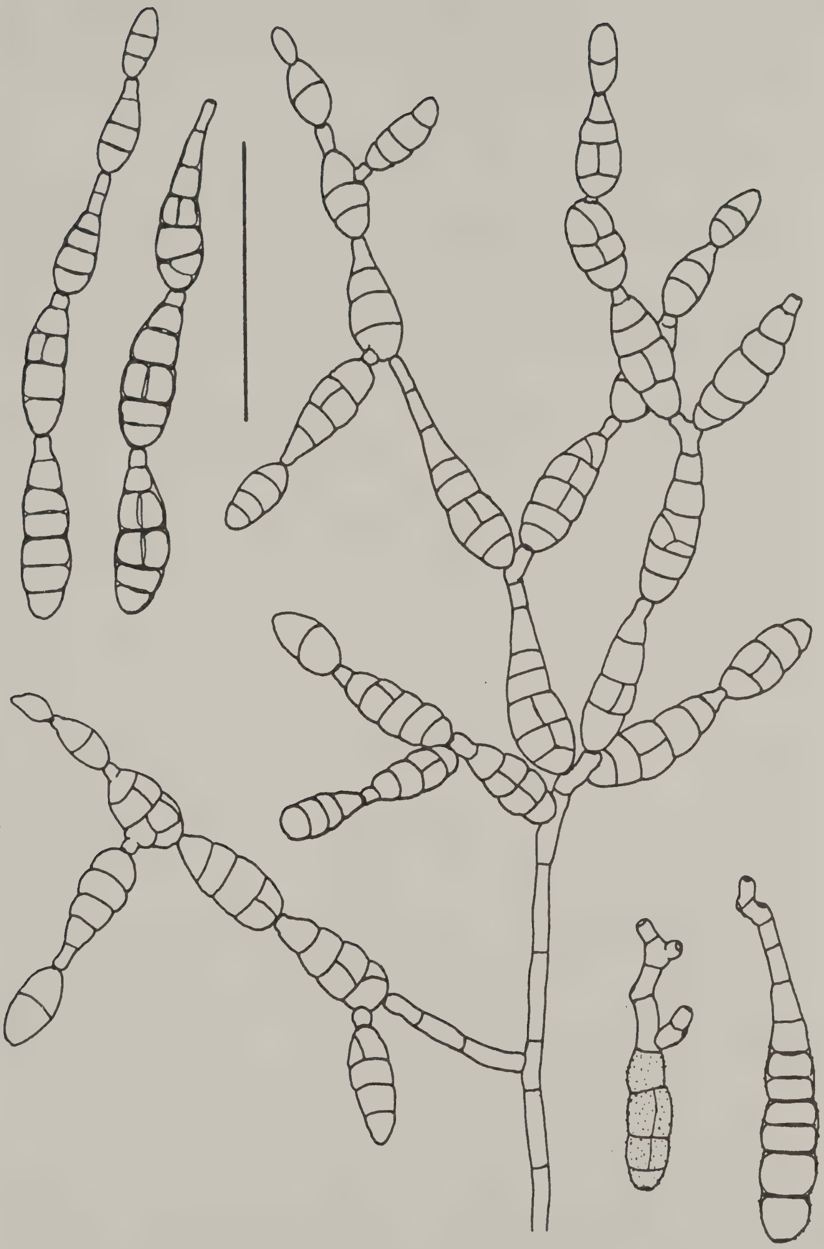

Fig. 165 contd. *Alternaria alternata*: sporulation clump and conidia from V-8; EGS 34-016. Bars = 50μm.

especially recommended, as it has been distributed for years to numerous research laboratories and individuals for comparison and standardization purposes. EGS 34-016 is used for the following description.

Description from PCA at 5-7d; Fig. 165

The sporulation pattern of *A. alternata* comprises a single suberect conidiophore and an apical cluster of branching chains of small conidia separated by short secondary conidiophores. The primary conidiophore is comparatively short, ca. 40-70 x 3-4μm; it remains simple or may become 1-3 branched or geniculate, with corresponding numbers of primary conidial chains. Branching within the primary chains is initiated when a few secondary conidiophores develop a series of conidiogenous loci or when secondary conidiophores arise from conidium body cells. The somewhat loose cluster of branching chains can be seen (at 50X) to consist of ca. 75-125 conidia. In older portions of a colony, the erect conidiophores and their sporulation heads present a picture of a solid turf of overlapping, interwoven conidial chains. In isolate EGS 34-016 and other isolates that are completely stable on PCA, there is no surface or aerial mass of non-sporulating mycelium, no growth of sterile hyphae, and no radial sectoring.

Single chains of conidia in the branching head (ignoring short lateral branches) have ca. 15-20 conidia. The first 1-2 conidia in a chain usually remain long-elliptical as they mature; conidia produced later in the chain become ovoid, ellipsoid, or subsphaeroid. Initial elliptical conidia are ca. 25-30(-40) x 5-9μm, with 4-7 transverse septa and a few or no longisepta; subsequent spores are ca. 7-25(-40) x 5-12μm, with 1-7 (very commonly 3) transepta and very few or no longisepta. The secondary conidiophore in *A. alternata* chains may be obsolescent, but almost always is a single cell ca. 2-3 x 3-5μm; occasionally it becomes 2-celled and, in the case of lateral conidiophores produced from conidium cells, up to ca. 30 x 3-4μm. Conidia become pale yellowish to golden brown after a few days in lactic acid mounts; wall ornamentation is densely punctulate in juvenile conidia, granular to variously verrucose as conidia mature.

Inclusive size ranges published for conidia of *A. alternata* are delusional and misleading unless they are related to specific growth conditions, colony age, chain length, and relative positions of conidia in the chain. Conidium size ranges divorced from the overall sporulation pattern that produced them are largely uninformative in modern *Alternaria* systematics.

237. ***Alternaria tenuissima*** (Fr.) Wiltshire (1933) Fig. 166

Background Information

Typical *A. tenuissima* is included here for comparison purposes because its sporulation in culture represents a pattern of chain formation found in some of the HST-producers. The basic pattern is of moderate-length to long chains of conidia, with or without lateral branches of conidia.

Alternaria species that display this sporulation habit in culture constitute a very high percentage of isolates that are encountered, from both saprophytic and parasitic situations. A few microscopically different species with the *A. tenuissima* sporulation pattern were illustrated in an earlier study of ex-citrus material (Simmons 1990, *Alt.T&V* 32-33). This *A. tenuissima* pattern of sporulation also has been discussed in several other contexts (Simmons 1981, *Alt.T&V* 6; 1993a, *Alt.T&V* 59; 1995, *Alt.T&V* 116-122). No species of the HST group discussed here is typical *A. tenuissima*.

Isolate Used for Description and Illustration

Isolate EGS 34-015 has been chosen to represent my concept of typical *A. tenuissima* as it develops under controlled conditions in axenic culture. The isolate and the neotype were described and illustrated in detail in *Alt.T&V* 116-117, figs. 94-95 (Simmons 1995).

Description from PCA at 5-7d; Fig. 166

The basic sporulation structure of *A. tenuissima* in both nature and axenic culture is simple, moderate-length chains of 5-10 or more conidia, a significant percentage of which are long-narrow. In culture the initial chain may remain simple, but commonly it produces one or a few lateral branches of a few conidia. In older cultures on PCA (10+d), chains may become 12-15 units long and typically have several conidial branches. The colony itself (viewed at 50X) advances from a stand of simple chains to a complex of long chains and their intertwined branches.

Initial conidia in a chain often have a narrow-tapered upper half. Conidia in the distal part of a chain usually are ovoid or ellipsoid without a narrow taper in the upper half; each conidium produces a short apical secondary conidiophore of 1-2 cells.

Narrowest conidia have only transverse septa. Wider conidia have both transepta (5-8) and a few longitudinal or

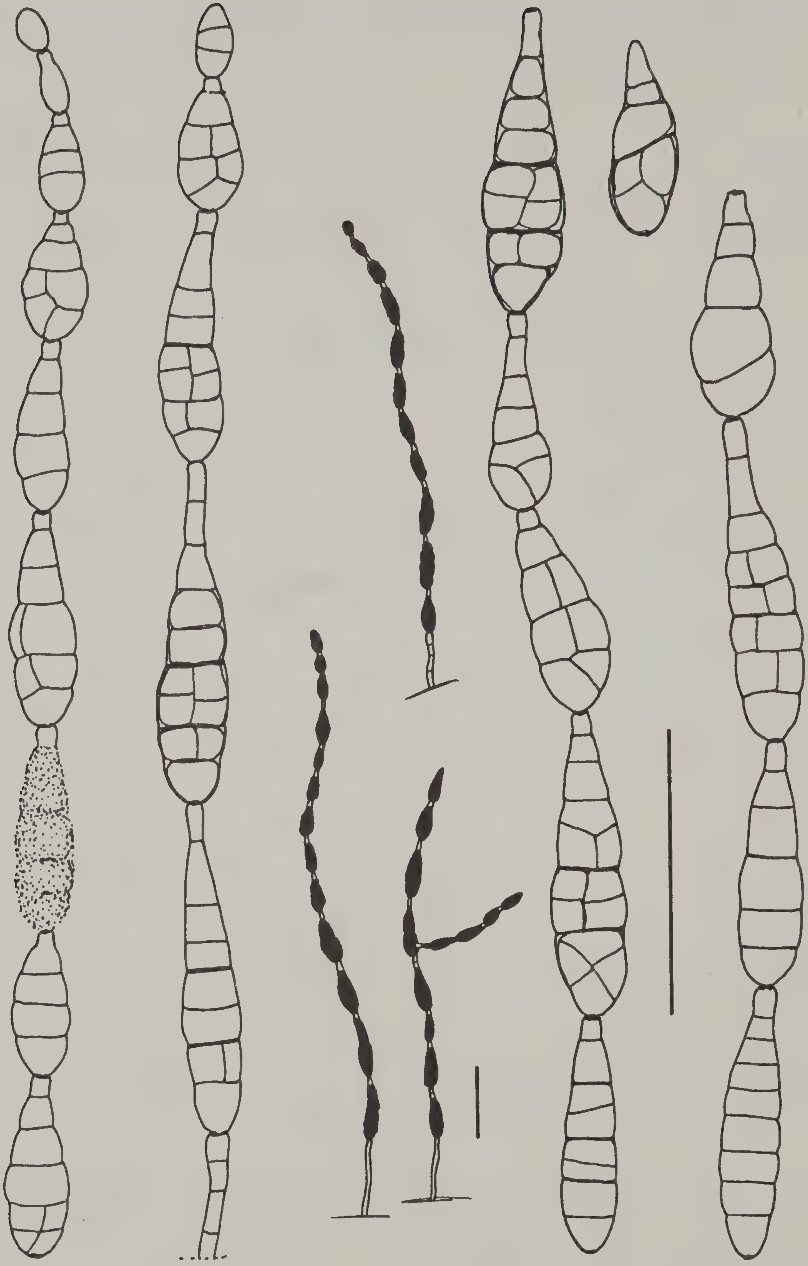

Fig. 166. *Alternaria tenuissima*: conidium chains and habit from PCA (2 chains at left) and V-8 (2 chains at right); EGS 34-015. Bars = 50μm

oblique septa in the broadest segments of the spore body. Only one of two of the largest conidia in a culture chain have a diagnostic median, somewhat constricting transverse septum, which is a striking feature of field conidia.

Conidia having only transepta reach a size range of ca. 32-45 x 11-13μm; ovoid muriformly septate conidia ca. 32-45 x 14-18μm; obclavate muriformly septate conidia ca. 40-60 x 16-18μm. Conidia are medium gold-brown and closely, obscurely to conspicuously punctulate.

238. ***Alternaria gaisen*** Nagano (1920) Fig. 167
= *Alternaria kikuchiana* Tanaka (1933)

Background Information

The history of naming and misnaming the *Alternaria* incitant of black spot disease of Japanese pear, *A. gaisen*, is detailed in *Alt.T&V* 63-70 (Simmons 1993b), along with an extensive description of a representative modern isolate (EGS 90-0512) from a selection of 19 morphologically identical isolates that proved to be toxigenic when tested against leaves of Japanese pear, *Pyrus pyrifolia* (N. L. Burm.) Nakai cv. Nijisseiki. The toxigenicity records of a selection of 100+ isolates in a pear black-spot study (Simmons and Roberts 1993) revealed that typical, toxigenic *A. gaisen* is very far from being the most common *Alternaria* species that develops on black-spot lesions under damp-chamber incubation and isolation procedures.

The great variety of sporulation patterns found among several hundred isolates used in the Simmons and Roberts study did not disguise, indeed it accentuated, the morphological uniqueness of *A. gaisen* and its identity with strains used in HST and molecular studies, specifically strain Tottori University 15A. The study was a forerunner of these current examinations and of the conclusion that neither *A. gaisen* nor any other of the HST-producing taxa treated here bears any significant resemblance to *A. alternata*.

Alternaria gaisen (as *A. kikuchiana*) was the first of the several HST-producing species studied intensively and published on extensively by S. Nishimura and his students and colleagues, mostly at Nagoya University and Tottori University in Japan. *A. gaisen* also was the first of these species to be misidentified as *A. alternata* on the basis of misapplication of information in Simmons (1967); *A. gaisen* therefore bears the burden of this misidentification being transferred and misapplied to other morphologically distinct HST-producing species.

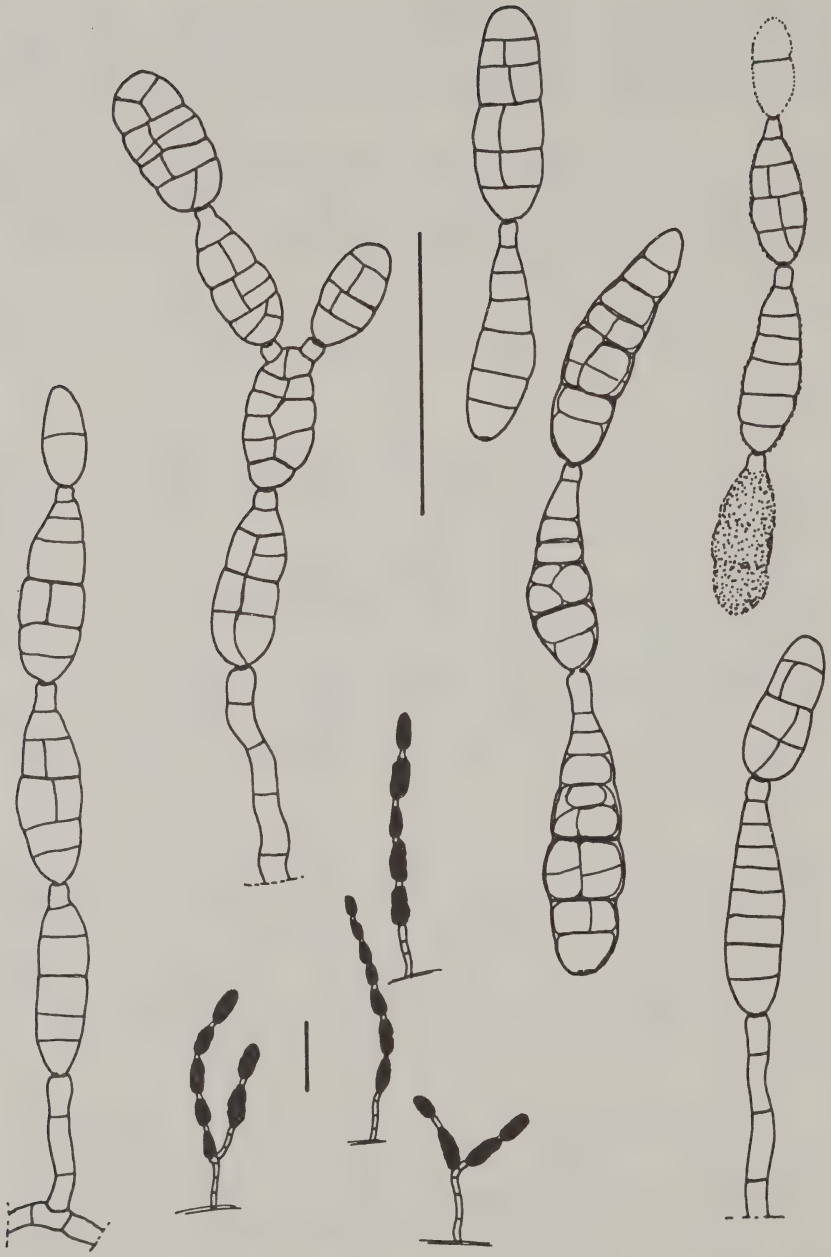

Fig. 167. *Alternaria gaisen*: conidium chains and sporulation habit from PCA (long chains at left) and V-8 (right of bar); EGS 90-0512. Bars = 50μm.

Isolate Used for Description and Illustration

Typical toxigenic *A. gaisen* is represented in my collection by numerous isolates that originated in Japan, Korea, and Taiwan. Strain EGS 90-0512 is a culturally stable isolate. It was isolated by me from a black-spot lesion of a Nijisseiki pear leaf collected in an experimental plot at Tottori University during a visit made 11 July 1990. Its sporulation morphology is identifiable with that of TU 15A, which often is cited as a standard strain in HST and allied research.

Description from PCA at 5-7d; Fig. 167

The extensive description of *A. gaisen* in Simmons (1993b) is condensed here for purposes of ready comparison.

Primary conidiophores of *A. gaisen* are solitary and uncrowded; they may be sporogenous when only 15-25μm long, but usually are (50-)80-150 x 3.0-4.5μm. Primary conidiophores usually are simple, but many at this age have 1-2 apical geniculations and conidiogenous sites. Conidial chains have 3-9 spores, usually remain unbranched, but may produce 1-2 short side branches from a conidium within the chain. Conidia are short to long ovoid or ellipsoid; they do not have a strongly tapered body or a true beak. They are 30-45(-55) x 13-15(-18)μm, medium yellow-brown, almost smooth to conspicuously verruculose overall, with 5-8 transverse and 0-1(-2) longitudinal septa in each transverse segment. Each conidium of a chain except the terminal one has an apical secondary conidiophore of 1 cell ca. 4.5 x 4.5μm; often the apical conidiogenous site is obsolescent, i.e., a simple pore; or uncommonly it is 2-3 cells and ca. 15 x 4.5μm.

Ageing cultures of *A. gaisen* (10d to 2wks or more), like those of other *Alternaria* species and great numbers of isolates of other genera, may have produced longer chains of conidia (to ca. 13 units), increased branching of conidiophores and chains, and noticeable amounts of non-sporulating hyphae. However, the morphology of 5-7d, active, culturally stable isolates and their sporulation elements under recommended conditions are as given above.

239. ***Alternaria limoniasperae*** Simmons (1999) Fig. 168

Background Information

As early as 1929, E. M. Doidge described a leafspot disease prevalent on rough lemon rootstock in South Africa. It could be induced experimentally by her isolates from rough lemon but not by isolates she tried from other citrus species. G. D. Ruehle (1937) reported similar negative results in Florida from cross inoculation with isolates from rough lemon lesions and from dry rot of citrus fruits. Similar results and conclusions, based on toxic filtrates applied to tangerine and rough lemon leaves, were reached by K. Kohmoto, R. P. Scheffer, and J. O. Whiteside (1979).

From the time of Doidge's initial study until the present, the incitant of *Alternaria* leaf spot of rough lemon has been recognized as host restricted; it is pathogenic, as far as is reported, only to *C. jambhiri* and to Rangpur lime, *C. limonia* Osbeck (Whiteside 1988).

Two of Whiteside's pathogenic isolates from rough lemon in Florida (PR 325 = EGS 39-185, PR 187 = EGS 39-187) figure prominently in HST studies (Kohmoto et al. 1991) and in studies of the phylogeny of HST strains (Kusaba and Tsuge 1995). The two strains are morphologically identical with 22 isolates from rough lemon discussed in support of the description of *A. limoniasperae* (Simmons 1999; *Alt.T&V* 226). Although the Whiteside strains are cited confusingly as "*Alternaria alternata* rough lemon pathotype" (Kohmoto et al. 1991) and "*Alternaria citri* rough lemon pathotype" (Kusaba and Tsuge 1995), neither they nor any of the more recently isolated rough lemon strains are identifiable as *A. alternata* or *A. citri*. The rough lemon isolates are readily distinguishable under controlled conditions of axenic culture and were used as bases for the description of *A. limoniasperae*.

Isolate Used for Description and Illustration

Isolate HC2-RLR-11s (EGS 45-100) was used in typification of *A. limoniasperae*. It is one of numerous identical isolates received in July 1997 from the University of Florida Citrus Research and Education Center, Lake Alfred, and used as part of the study of 135 ex-citrus strains recorded in *Alt.T&V* 226-235 (Simmons 1999).

Unlike other species in this group of pathogens, *A. limoniasperae* can be identified most readily from sporulation produced on poorly nutritive Hay-decoction agar. The following description characterizes the species on both PCA (for comparison purposes) and on Hay.

Description from PCA and Hay at 5-7d; Fig. 168

Primary conidiophores are up to ca. 100 x 5-6μm in light-exposed areas, or up to 0.5-1.0mm x 5μm in dark-phase zones of growth.

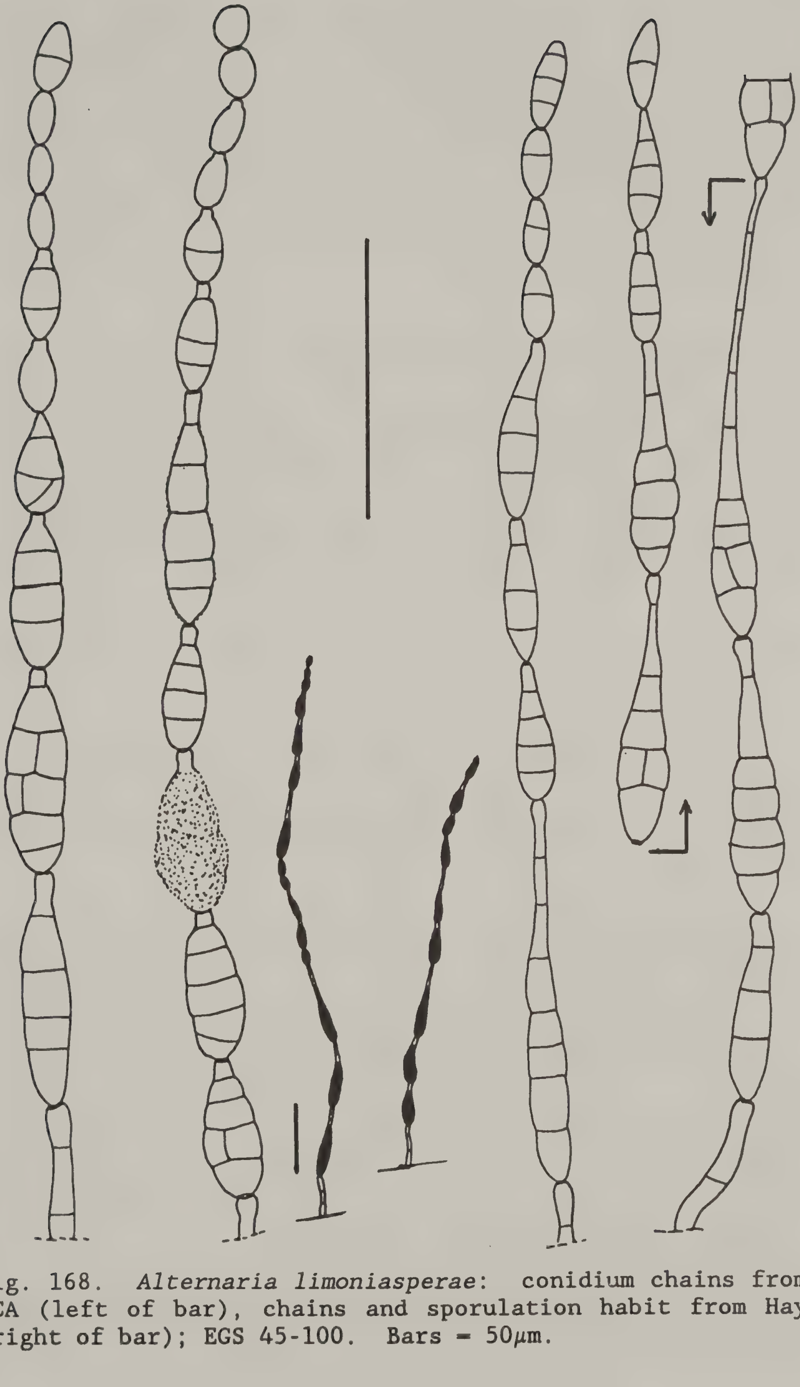

Fig. 168. *Alternaria limoniasperae*: conidium chains from PCA (left of bar), chains and sporulation habit from Hay (right of bar); EGS 45-100. Bars = 50μm.

Conidial chains on PCA elongate from 4-6 spores (at 2d) to 10-12 units (5d) to ca. 20 units (7-10d) and commonly have 1-4 lateral branches of 4-10 spores each. Conidia are narrow-ellipsoid 30-50 x 8-10μm, or ovoid 20-35 x 8-12μm, in the early lower portion of the chain but become ovoid and smaller (8-12 x 4-8μm) in the terminal portion of the chain. Secondary conidiophores are 2-4 x 2-3μm or obsolescent to the form of a simple conidiogenous pore; have 1-4 transepta and 1(-2) longisepta in relatively few conidia of a chain, and are medium gold-brown with punctulate to punctate ornamentation overall.

Conidial chains on Hay are very long and narrow, with 10-12 spores (at 4d) to 10-30 units as the colony ages (7-10d). Conidia in early growth are ca. 25-40 x 7-11μm and have apical secondary conidiophores that are mostly 2-4 x 2-3μm but also in some conidia are strikingly 25-50 x 2-3μm. This narrow apical extension, often 1-2 times as long as the spore body, gives the chains (at 50X) an elongate, fragile appearance that is diagnostic among the species discussed here. The type description from Hay culture is much more detailed in Simmons (1999).

240. *Alternaria toxicogenica* Simmons (1999) Fig. 169

Background Information

Among the several ex-citrus isolates shared with me in 1990 by J. O. Whiteside is his isolate PR 320 from Dancy tangerine in Florida; the isolate is morphologically unique among the 80-90 tangerine-related strains studied for *Alt.T&V* 226-235 (Simmons 1999).

It is remarkable that this single isolate was the basis for early reports of the "tangerine pathotype" of "*Alternaria alternata*" (Kohmoto et al. 1991), for if other isolates of the same species are known, they are not identified as equivalent in the HST literature. It is equally remarkable that—given the exquisitely detailed attention paid to filter pore size, reagent grade, name of equipment manufacturers, etc., etc., in some of the HST publications—rarely is the absolutely essential toxin-producing *Alternaria* isolate given a unique strain number or is vouchered in an identified culture collection. The likelihood is great that several different *Alternaria* species are submerged in an ambiguous "tangerine pathotype"; see Simmons (1999).

The morphological range of different species able to induce the pathology known as "*Alternaria* brown spot of mandarins," (Simmons 1999, *Alt.T&V* 228-235) suggests that the

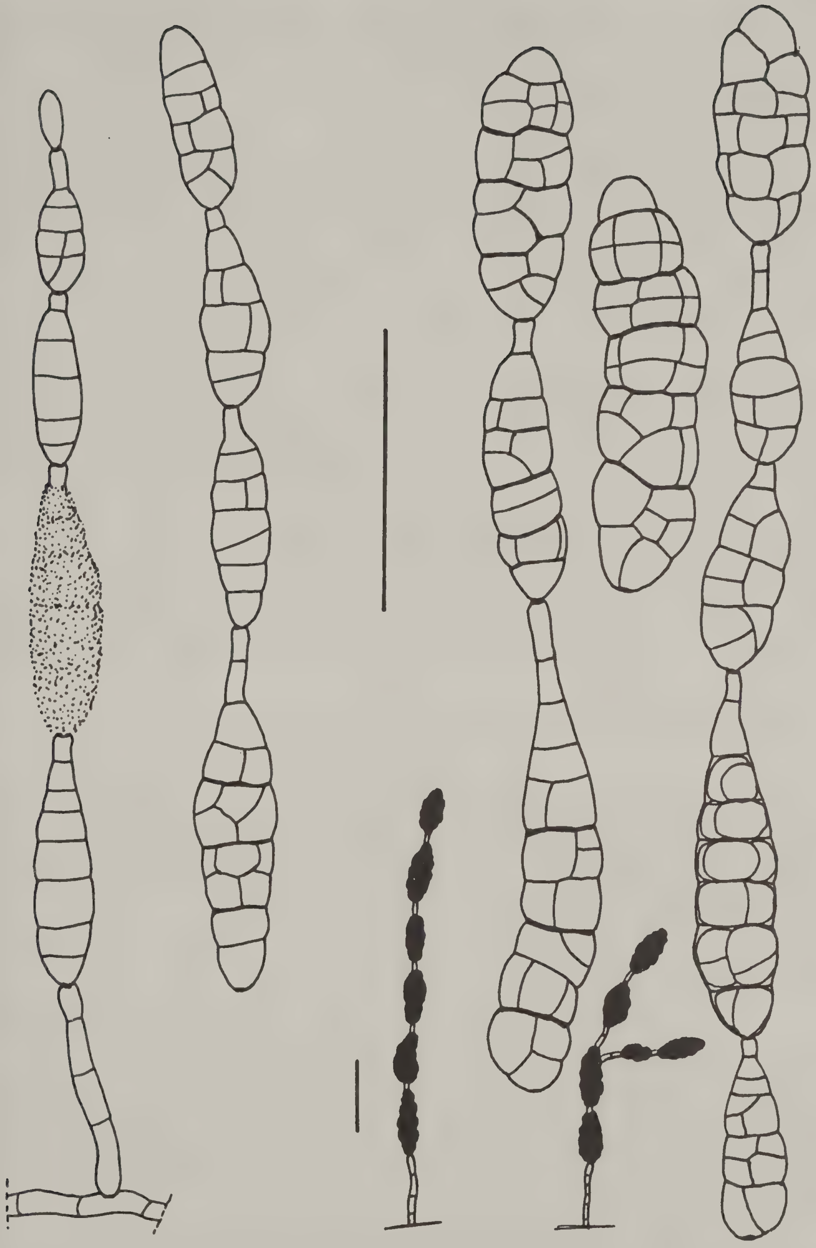

Fig. 169. *Alternaria toxicogenica*: conidium chains and sporulation habit from PCA (left of bar) and V-8 (right of bar); EGS 39-181. Bars = 50µm.

nine host-specific toxins reported from tangerine-related hosts (Montemurro and Visconti 1992) are likely to be products of several distinct *Alternaria* species—quite certainly more than one.

The likelihood that the "tangerine pathotype" concept has any taxonomic or identification value in *Alternaria* is negligible. An isolate of *Alternaria* from *Brassica sinensis* [?= *B. chinensis* L.] in central Germany is reported to produce ACTG [*Alternaria citri* tangerine pathotype] toxins D and E. These results indicate that neither toxin can now be considered to be host specific (Liebermann, et al. 1994).

Research at Tottori University informs us, in a similar vein, that some *Alternaria* isolates from Emperor mandarin (Itoh et al. 1993) and Australia tangerine (Fujii et al. 1998) produce toxins that are pathogenically active against not only the original citrus hosts but also strongly against one of the most thoroughly studied of the non-*Citrus* hosts, Japanese pear cv. Nijisseiki (*Pyrus pyrifolia* (N. L. Burm.) Nakai). The concept of the HST "tangerine pathotype" loses force when *Alternaria* isolates from citrus are equally toxic to Japanese pear, and when isolates from *Brassica* yield products that are chemically identical with toxins described as HSTs for *Citrus*.

Isolate Used for Description and Illustration

Whiteside's isolate PR 320 has no morphologic twin among the 80-90 stable isolates from brown-spot lesions examined from sources in Colombia, Israel, South Africa, Turkey, and Florida, U.S.A. (Simmons 1999). Its short-chain, large-conidium sporulation apparatus is presented from PCA for comparison purposes and V-8 for emphasis.

Description from PCA and V-8 at 5-7d; Fig. 169

Primary conidiophores are 50-100 x 4-5μm; they commonly become 1-2 geniculate near the apex and generate 2-3 simple chains of conidia. Chains of conidia are short, 2-6(-10); occasional branches usually have only 1-2 spores. Simple chains are the rule, but chain branching and conidiogenous conidiophore extensions may generate small clumps of 15-20+ conidia.

Conidia in young chains are long-ovoid, ca. 30-50 x 11-16μm; in older chains, broad-ellipsoid, ca. 25-45 x 15-20μm; have a 1-2 cell secondary conidiophore ca. 3-12 x 3μm; usually have a median constricting septum and 2-6 additional transepta, with secondary oblique and longitudinal septa inserted rather irregularly; are medium dull gold-brown, smooth to punctulate or strongly punctate.

Alternaria toxicogenica was typified from culture on V-8, which produces conidia notably larger than on PCA. Conidia from V-8 are broad-ovoid or broad-ellipsoid, 35-70 x 15-18μm; usually have 7-9 transverse septa, of which 3-5 are primary and constricting, and several oblique and longitudinal secondary septa.

241. ***Alternaria longipes*** (E. & E.) Mason (1928) Fig. 170

Background Information

The *Alternaria* species that incites brown leafspot of tobacco (*Nicotiana tabacum* L.), paralleled with its demonstrated toxigenicity, is best understood taxonomically through a review of three sources of information:

- first, a study of its type specimen (Simmons 1981; EGS 03-094, NY as *Macrosporium longipes* Ell. & Ev.), and comparison of conidia from a modern field collection (EGS 33-095) and from a representative, very virulent isolate (EGS 30-033 = Stavely A-5);

- second, a presentation of culture characters and excellent illustrations of typical conidia (Sobers and Doupnik 1969);

- third, a series of blind comparisons in the early 1970s, designed to challenge my ability to select morphologically typical *A. longipes* isolates that would match toxicity data derived independently from the same isolates at tobacco research institutions in North Carolina. The positive results of this cooperative study were never published, to my disappointment and chagrin, but are summarized here as evidence that *A. longipes* is both morphologically distinguishable and predictably toxigenic.

Referring to unpublished results, even from a time-consuming effort that I have not mentioned in earlier discussion of *A. longipes* (Simmons 1981, *Alt.T&V* 6), is not a satisfying way to present a case. However, in assembling literature for these present notes, I find that my unpublished opinions on some of the pertinent isolates from tobacco have been discussed in print (e.g., Spurr 1977), so my restraint has evaporated, as indicated in the following six paragraphs.

In 1971-1972, ninety *Alternaria* isolates were sent to me by tobacco pathologists associated with North Carolina State University, Raleigh, with the university's Agricultural Experiment Station, and with laboratories of the ARS/USDA, Oxford, North Carolina. No toxicity data were included; no host/substrate information was given, as I had asked for isolates to come from miscellaneous sources in addition to tobacco. (The sources later were revealed to be tobacco, apple, wheat, tomato, and popcorn.)

The narrowest interpretation of typical *A. longipes* growth and sporulation morphology under controlled culture conditions was used (Simmons 1981, Sobers and Doupnik 1969). Culturally unstable isolates, of which there were several, were ignored in initial decisions.

Nine of the first 70 isolates studied were identified morphologically by me as *A. longipes*; when matched with toxicity data developed in North Carolina (method of Spurr 1973), these 9 represented two-thirds of the 14 most virulent isolates known to the North Carolina laboratories. Noteworthy from the morphotaxonomic viewpoint, as it should be from the pathology angle: in no instance was any one of the 55+ low-virulence or non-toxigenic isolates mistakenly identified as *A. longipes*.

Distinguishing *A. longipes* from other small-spored *Alternaria* species commonly found on tobacco leaves, often even in brown-leafspot lesions, has a high success rate when culturally stable isolates are studied under specific controlled conditions. Prediction of toxigenicity by morphologically identified isolates has the same high rate of success.

In spite of this high level of predictability of an identifiable *A. longipes*/tobacco-pathology relationship, staff pressures among university and USDA pathologists led to a name choice, *A. alternata*, that would cause the least friction on site. In choosing the name *A. alternata* to represent the pathogen causing brown leafspot of tobacco, the classification lumpers overrode the results of a well-controlled pathogen/toxicity identification effort; the results were not published. It is no longer important to detail the names of supporters and detractors of the decision among tobacco pathologists involved, particularly as the decision to settle on *A. alternata* in this instance was reported to be by an internal review committee, anonymous to me.

The use of *A. alternata* for the tobacco pathogen began with G. B. Lucas (1971), who felt that he could not distinguish the tobacco pathogen from non-toxic *Alternaria* isolates

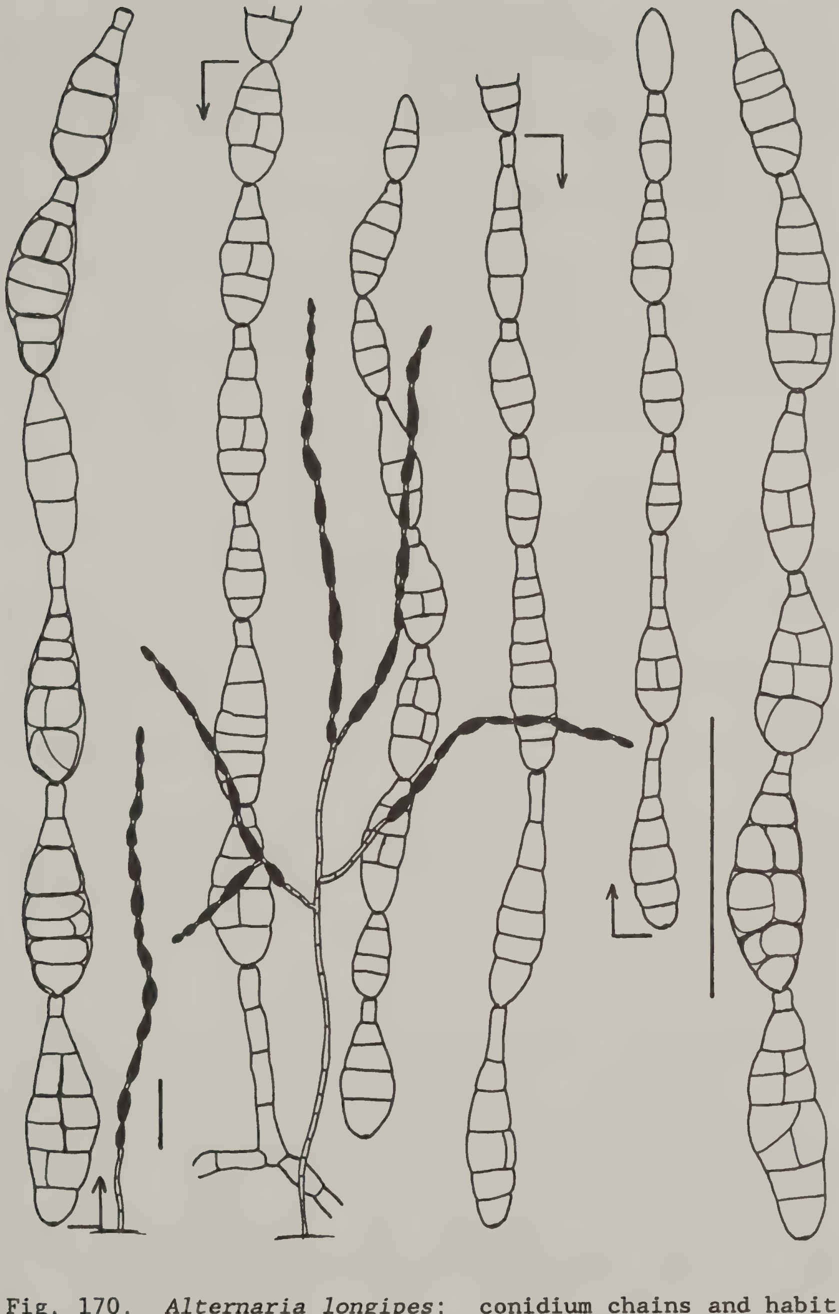

Fig. 170. *Alternaria longipes*: conidium chains and habit from PCA (left of bar) and V-8 (right of bar); EGS 30-033.

from tobacco, a view that I challenged in detail when he asked me to review his prepublication manuscript. The usage is found subsequently in Spurr (1977), Spurr and Main (1974), and Spurr and Welty (1975), with its identification error extending into the HST literature as the "*A. alternata* tobacco pathotype."

Isolate Used for Description and Illustration

An excellent representative of *A. longipes*, EGS 30-033, has remained culturally and morphologically stable in my hands since its receipt in 1971 as Stavely A-5. At that time it still was "as pathogenic as when originally isolated" in North Carolina in 1967 (*in litt.* J. R. Stavely, ARS/USDA, Beltsville, Maryland). Virulence was 4.5 (on a scale of 1.0-5.0), i.e., "very virulent" in the comparisons begun in 1971-1972.

Several other morphologically typical and experimentally virulent strains are listed in Simmons (1981, *Alt.T&V* 6). One isolate of that list should be deleted, as it is neither typical nor toxigenic, viz., EGS 16-135 ex G. B. Lucas R-66 = ATCC 15052 = CBS 620.83.

A major factor contributing to misidentification of *A. longipes* is that, judging from conserved and contributed specimens, *A. longipes* often sporulates poorly on typical leaf lesions, whereas other small-spored *Alternaria* species often are present in abundance and, predictably, are more likely to be noted and isolated than is the primary pathogen. Confusion of this sort has led to large quantities of discriminating work in phytopathology and biochemistry, not to mention taxonomy, being dumped in the literature under *A. alternata*, a demonstrably incorrect name for the *Alternaria* that causes the brown leafspot disease of tobacco.

Description from PCA at ca. 7d; Fig. 170

The sporulation pattern of *A. longipes* often is of long simple chains of 10-15+ conidia without lateral branches or with 1-3 branches of 1-7 conidia; supporting primary conidiophores are up to ca. 60µm long. Equally prevalent and characteristic are long primary conidiophores, up to ca. 500µm, with 1-4 horizontal or ascending branches, each of which bears a simple or occasionally branching chain of conidia. Conidial chains are very abundant but, at 50X, appear distinct and uncrowded, never massed into a dense head of conidia.

Conidia are narrow-ovoid or ellipsoid, with a general appearance of simplicity in that longitudinal septa are lacking or uncommon in large numbers of the narrow conidia. The body of conidia produced on PCA is 12-35(-48) x 5-12(-14)μm, with 3-8 transverse septa and no longisepta or only 1(-2) in 1-2 cells of narrower conidia or a few more in the broader conidia; pigmentation of fresh conidia is a dull olive-brown, which changes to a distinctive tawny brown within a few days in lactic acid mounts. Conidia are smooth or have punctate ornamentation overall. All but bluntly rounded terminal conidia have an apical secondary conidiophore, which usually is 1-celled, 3 x 3μm; longer, septate secondary conidiophores are common, up to ca. 30 x 3μm, and are obvious reproductive outgrowths, not a conidium beak integral with the spore body.

242. ***Alternaria mali*** Roberts (1914) Fig. 171

Background Information

J. W. Roberts (1914) initially described an *Alternaria* isolated from apple fruit; it was able to cause a rapid rotting of ripe apples as well as to enlarge dead spots of apple leaves. The typifying description of 20-25 words was greatly expanded in a second publication (Roberts 1924), where, after additional experimentation, he reported details of conidium characters, especially size ranges related to degree of septation.

Whether or not the species Roberts studied is related to the disease now generally known as *Alternaria* blotch of apple, it can be demonstrated that his *A. mali* is a fungus morphologically different than the one that figures prominently in HST-related literature (Tottori University 0-159).

Alternaria mali also differs morphologically at the species level from isolates received by me in 1984 from two research institutions in Japan, the Aomori Apple Experimental Station (*A. mali* Ao-4, Ao-5 = EGS 37-128/-129) and Hirosaki University (AKI-3 = EGS 37-131); from isolates made by me from typical apple blotch leaf lesions, indicated as such by resident staff at two host institutions in Japan, Nagano Nanshin Agricultural Experimental Station (EGS 90-053/-054/-055) and Fukushima Fruit Experimental Station (EGS 90-056/-057/-058/-059); and from my isolates from typical apple blotch specimens received in 1991 from T. B. Sutton (North Carolina State University, Raleigh) in connection with work reported by Filadić and Sutton (1991) (EGS 91-0242/-0243/-0281/-0282).

So many different and definable sporulation patterns and conidium morphologies exist among isolates cited above that at least one hypothesis requires consideration, i.e., that the pathology known as "*Alternaria* blotch of apple," like that of "brown spot of tangerine," can be induced by several different species of *Alternaria*; and as a corollary, that if a single identifiable host-specific toxin is a participant in the disease, then that HST is produced by at least those several species. Or are several different toxins involved in a non-specific pathology?

In any case, the so-called "*A. alternata* apple pathotype" as represented by isolate TU 0-159 is neither *A. alternata* nor *A. mali*. Characterizing and differentiating the several ex-apple blotch isolates, including TU 0-159, is a separate project with, it is hoped, molecular-data input.

Isolate Used for Description and Illustration

The uniqueness of *A. mali*, in comparison with all of the isolates listed above, is readily demonstrated by examining the representative culture CBS 106.24, which Roberts deposited in 1924 with the Centraalbureau voor Schimmelcultures, Baarn, The Netherlands, to voucher his studies. The isolate remains in good condition, with stable growth and excellent sporulation under my culture conditions (EGS 38-029). It is an absurdity that this single isolate with authentic relationship to *A. mali* Roberts is hidden in catalogues as *A. alternata* (CBS 106.24, ATCC 13963), whereas Tottori University 0-159, incorrectly identified, is listed as *A. mali* (ATCC 42096). Further, it is phytopathologically bizarre that so much apple blotch information is attributed to so many different misidentified *Alternaria* isolates.

Description from PCA at ca.5-7d.; Fig. 171

Primary conidiophores of *A. mali* are, for the most part, relatively short, 50-100μm x 2-3μm, when they emerge directly from submerged or surface hyphae. They typically are simple, sometimes 1-branched. Long horizontal hyphae can be seen winding among the erect conidiophores; these pale hyphae produce darker, erect conidiogenous branches at various intervals.

The 50X aspect is of long narrow conidial chains, suberect at various angles, so that they appear to intermingle and lean on each other. Chains usually consist of 10-15(-20) spores. Branching within the chain is not a common feature; branches, when present, produce only a few conidia.

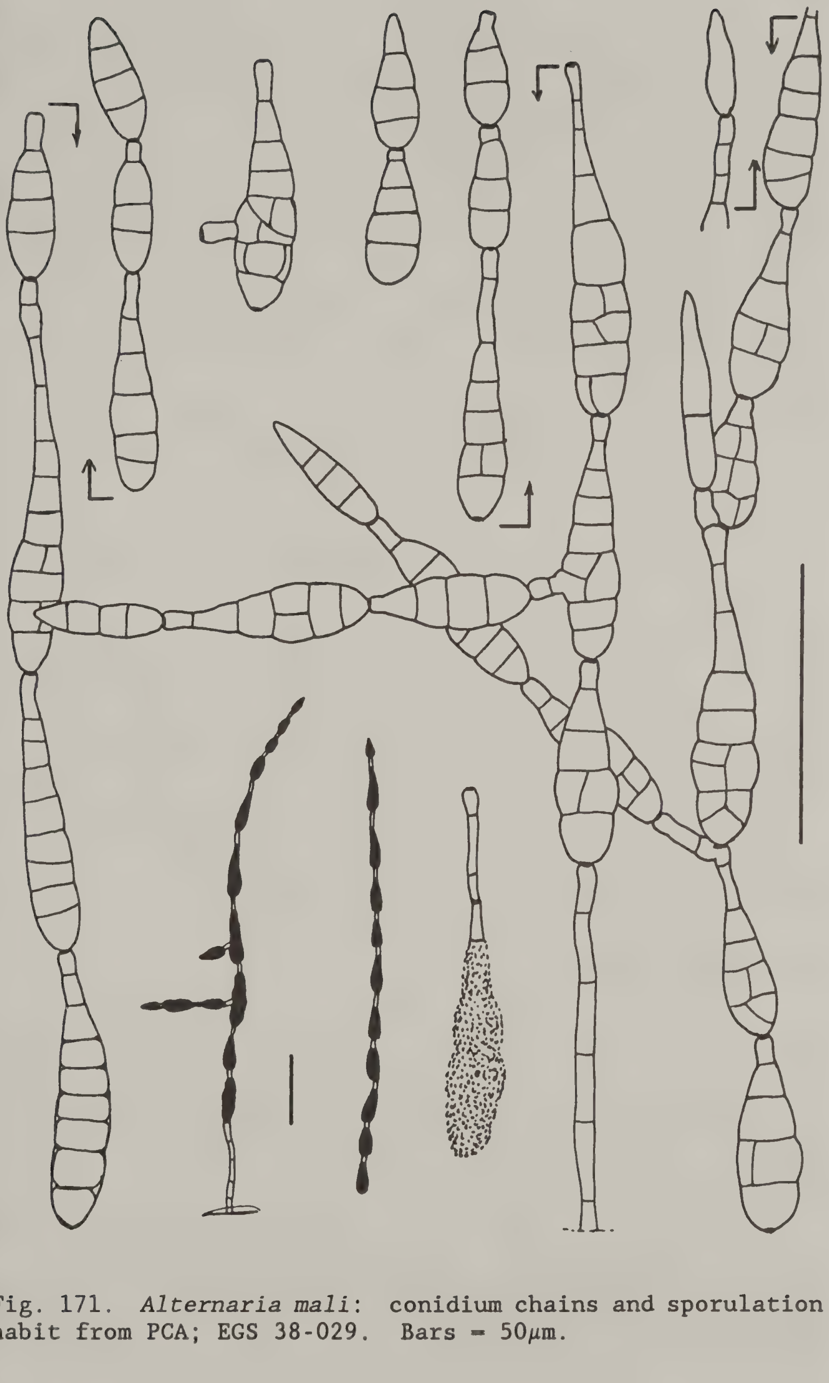

Fig. 171. *Alternaria mali*: conidium chains and sporulation habit from PCA; EGS 38-029. Bars = 50μm.

Most conidia, except for young ovoid units near the tip of the chain, are narrow-ovoid, i.e., with a broader lower half that gradually narrows upward, often wedge-like, to the 2-3μm width of the secondary conidiophore that usually is present. The body of narrow-ovoid conidia is 30-45(-50) x 5-8(-10)μm, with 5-8 transepta and usually no longisepta or, at most, only 1 longiseptum in 1-2 transverse segments of the broadest conidia of a chain. Conidium color is a dull dilute brown. Conidium walls are conspicuously ornamented overall, from punctate to verrucose. Apical secondary conidiophores may be obsolescent, but usually range from 1-celled 2-3 x 3-5μm to elongate 10-30+ x 2-3μm. A secondary apical conidiophore uncommonly generates a geniculate conidiogenous branch within the chain.

243. ***Alternaria arborescens*** Simmons, sp. nov. Fig. 172
= *Alternaria alternata* f.sp. *lycopersici* (Grogan et al. 1975)
Etym.: L. (part.), *arbor* + *escens*, becoming tree-like

*Ex cultura in agaro PCA descripta. Conidiophora suberecta, primigena ca. 200-300 x 3-4μm (in luce), secunda ad 800-1000+μm (in statu obscuro). Ordinatio sporulationis aspectu arboreo, constatae ex conidiophoris secundariis brevibus et catenis conidiorum ramosis; catenae conidiorum singulae constate ex conidiis 2-6; capitulum sporulationis constate ex conidiis 50-125. Conidia ovoidea vel ellipsoidea, 12-30(-42) x 7-11μm, conidiophoris secundariis 3-20(-30) x 3μm, 1-4 transverse, 0-2 longitudinaliter septata, dilute brunnea, manifeste punctata vel verruculosa. Habitatio typi in laesione cauline Lycopersicon esculentum Mill. Typus: partes ex cultura EGS 39-128 (AS27-3pz, a D. Gilchrist accept. 24.v.1990) desiccatae et in **BPI** (holotypus) et saltem in **IMI** (isotypus) conservandae.*

Background Information

A stem canker disease of tomato (*Lycopersicon esculentum* Mill.) was attributed to pathogenic isolates referred to as *A. alternata* f.sp. *lycopersici* by the initial investigators at the University of California, Davis (Grogan et al. 1975), and subsequently as *A. alternata* "tomato pathotype" in the HST literature (Nishimura and Nakatsuka 1989, Kusaba and Tsuge 1995, and others).

The initial publication included photographs of conidia from several substrates: a naturally infected stem, sterilized tomato stems in vitro, cornmeal agar, and a stem canker produced by inoculation methods. It also reported comparisons of conidium measurements of the stem-canker fungus with those of *A. alternata* as given for a few spores of the neotype specimen by Simmons (1967) and for synonymic *A. tenuis* by Groves and Skolko (1944) and by Neergaard (1945). The range of substrates upon which these measurement comparisons were based included natural field stems, sterilized stems, cornmeal agar, an unidentified stem at least 150 years old (the neotype), malt agar (as used by Groves and Skolko), and the "2-4 media" used by Neergaard (malt agar; a standard nutrient agar of salts, dextrose and asparagine; and usually seedlings of tomato and lettuce). The conidium sizes of the tomato pathogen appeared close enough to those given by the authors cited for *A. alternata* (= *A. tenuis*) that the authors of the f.sp. *lycopersici* opted for a species determination *A. alternata*. Given the very obvious size-range differences, as illustrated and reported from the considerable variety of substrates used by the several authors, I suspect that the tomato pathogen investigators themselves, 20 years later, might raise an eyebrow, as do I, about the validity of such a comparison and its usefulness today in a species decision.

Isolate Used for Description and Illustration

The isolate of *A. arborescens* (= *A. alternata* f.sp. *lycopersici*) cited in the molecular studies of Kusaba and Tsuge (1995) is credited as a Tottori University accession As-27. My own culture of the species arrived in 1990 directly from the originating laboratory at the University of California, Davis, as strain AS27-3pz (EGS 39-128). Another culture arrived recently for comparison as strain AS27-3iz (EGS 46-187, from University of California, Davis, via D. J. Vakalounakis, Heraklio, Greece).

I have not seen the Tottori University strain; my strain and the one via Vakalounakis are morphologically identical. Only strain EGS 39-128 is discussed here; it was received in response to a request for a typical representative of the tomato stem-canker pathogen.

Description from PCA at ca. 5-7d.; Fig. 172

Long, well-defined primary conidiophores of *A. arborescens* characteristically bear a few terminal and subterminal branches. Each conidiophore branch bears a branching chain of conidia, giving a relatively tall, three-dimensionally arborescent appearance to the suberect system. Primary conidiophores of moderate length are ca. 200-300 x 3-4μm when they produce a terminal cluster of branches and conidial chains. Other diagnostic, very long conidiophores commonly are up to ca. 800-900μm long before subterminal branches appear, and 1.0mm or longer when the full terminal

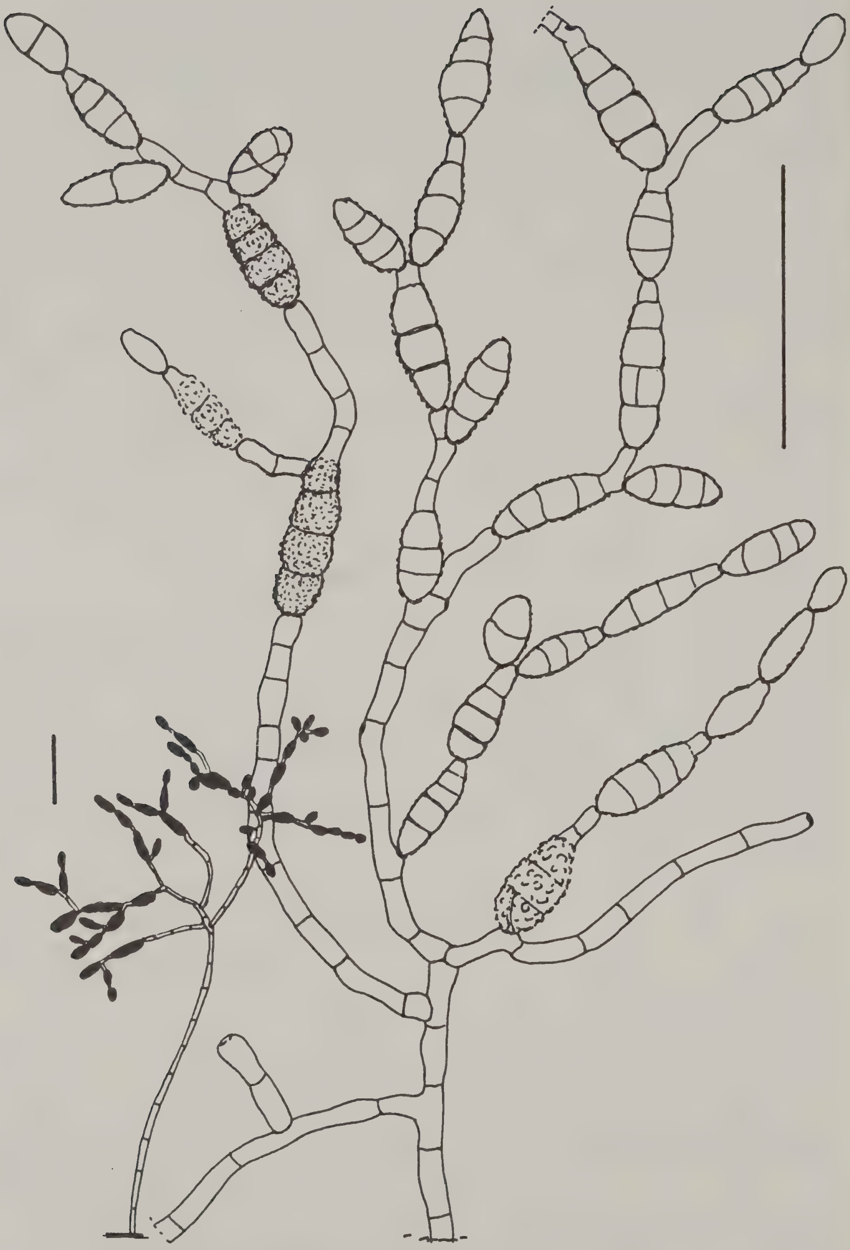

Fig. 172. *Alternaria arborescens*: portion of the terminal sporulation clump of long conidiophore habit shown in Fig. 172 contd., and habit of a short-conidiophore sporulation clump; EGS 39-128. Bars = 50μm.

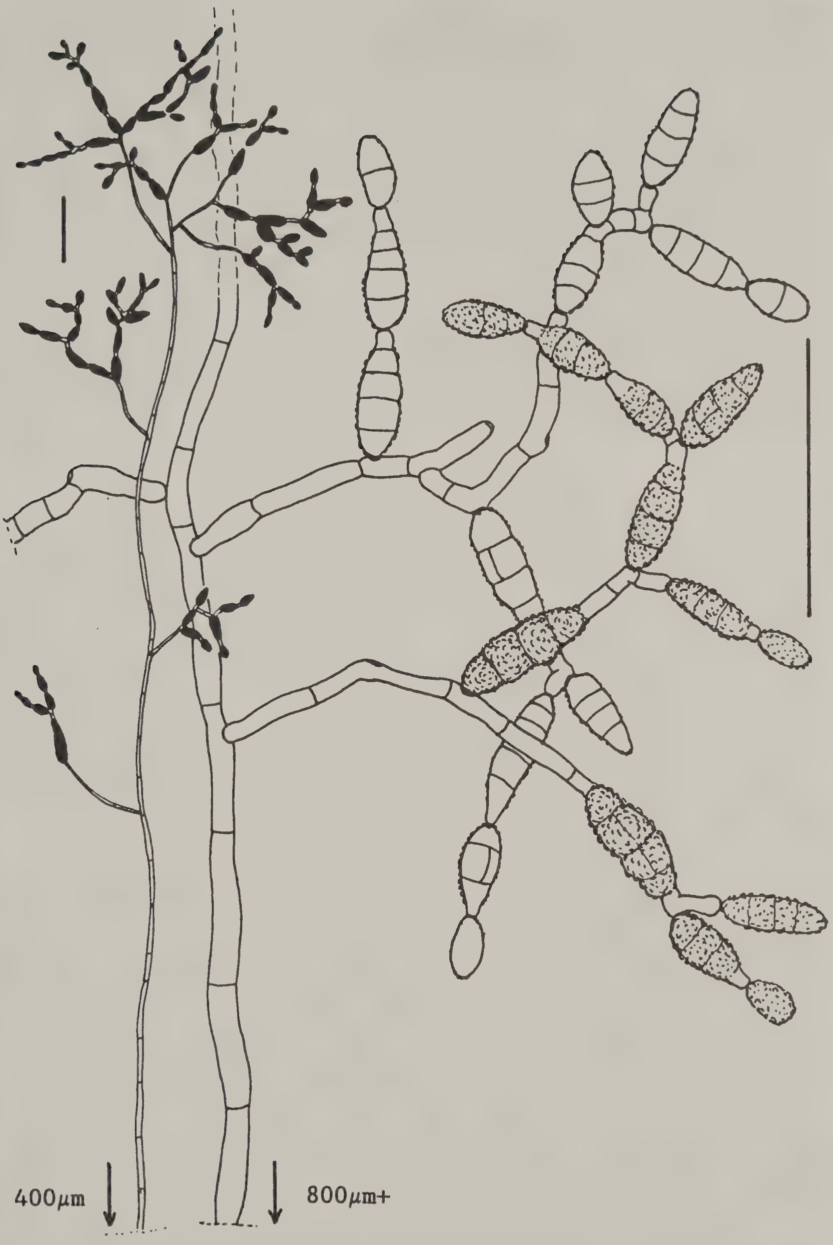

Fig. 172 contd. *Alternaria arborescens*: portion of a subterminal clump of long conidiophore habit, and terminal portion of long conidiophore habit; EGS 39-128. Bars = 50μm.

head of branches and conidia is produced. The sporulation head commonly consists of ca. 50-125 conidia. Major conidial chains are 2-6 units long and typically produce branches of 1-5 conidia.

Conidia are short-ovoid or ellipsoid. The body of conidia produced on PCA is 12-30(-42) x 7-11μm, with 1-4 transepta and no longisepta, or 1-2 longitudinal or oblique septa in a small percentage of the spore population. Color is a dilute tan to brown, with walls and septa contrastingly darker. All but youngest conidia are prominently punctate to verrucose. All but terminal conidia have apical secondary conidiophores, which sometimes are so short as to be obsolescent but usually are 3-20(-30) x ca. 3μm. These secondary conidiophores may be simple or commonly are 1-3 geniculate, with a conidiogenous site at each bend.

Discussion and Conclusions

The introduction and the conclusion of this study have the same bottom line: six named *Alternaria* species represented among major "host-specific toxin producers" are readily identifiable morphologically and are distinguishable from typical *A. alternata*. The equipment and conditions needed for checking this statement are modest, and certainly they are available to any phytopathology laboratory capable of performing HST research. The only extra ingredient required is a willingness to examine the identification premise with an enquiring mind, using culture and microscope techniques that are as current in mycotaxonomy as are HST or other techniques in mycology and phytopathology.

The approach in the text of these notes has been strictly morphologic, because the original decision and continuing practice of treating several HST-producing species as "pathotype" entities under a single *Alternaria* species name were and remain simple mistakes in interpreting and applying morphology data. The originators of the "collective species" concept in HST work decided that they could not distinguish conidia of their isolates from conidia of the field type of *A. alternata* by measuring conidia. But they and their copiers in the discipline have not re-evaluated their position, even when species of quite remarkable morphological differences have been added to the HST stable.

The phrase that appears in some form in most HST papers for the several toxigenic isolates—"*Alternaria alternata* collective species"—obviously has been repeated by rote from one paper to another. No pertinent publication, and certainly no paper in the HST field, presents morphologic data comparisons of any sort—a totally unacceptable procedure when taxonomic identity statements are being made.

Each of the species treated here has its own morphologic identity, as illustrated (Fig. 173). The HST-producers, as a group, have no classification affinity other than as small-spored taxa of *Alternaria*. The only relational character that has held them together in the HST literature is production of toxins, which themselves are of considerable chemical diversity.

The molecular and genetic information published on *Alternaria* HST isolates thus far is not extensive. I have been privileged to see molecular and secondary metabolite data that currently are being produced for some of the taxa. But these data are not mine to discuss before they are published.

In the molecular field, Kusaba and Tsuge (1995) have published a study of *Alternaria* species based on variation in internal transcribed spacers of ribosomal DNA. Their experimental material included isolates already discussed in the HST literature, as well as strains identified as *A. alternata* and seven isolates of relatively large-spored *Alternaria* species that they felt would not be misidentified as *A. alternata*. They concluded that their data show that the several HST taxa cannot be distinguished from *A. alternata* on an ITS-sequence comparison basis, and that the large-spored species cluster separately.

The logic of my reaction to the Kusaba and Tsuge interpretation is that the gene they have studied has insufficient relationship to morphology to support their one-species conclusion for HST taxa. The phylogeny of this gene is not necessarily congruent with a phylogeny of the several species that could be derived from a more comprehensive set of genetic data. I suggest that their two major clusters of data can be interpreted as valid at a subgeneric level of *Alternaria* classification—but not conclusively, and certainly not usefully, at the species level.

More recently, D. Cooke and colleagues (Cooke et al. 1998) analyzed intra- and interspecific variation in 13 species of *Alternaria* by random amplification of polymorphic DNA/polymerase chain reaction techniques. Their selection of species to be studied was based on morphologic groupings proposed by Simmons (1992). I am familiar with several of the isolates they used.

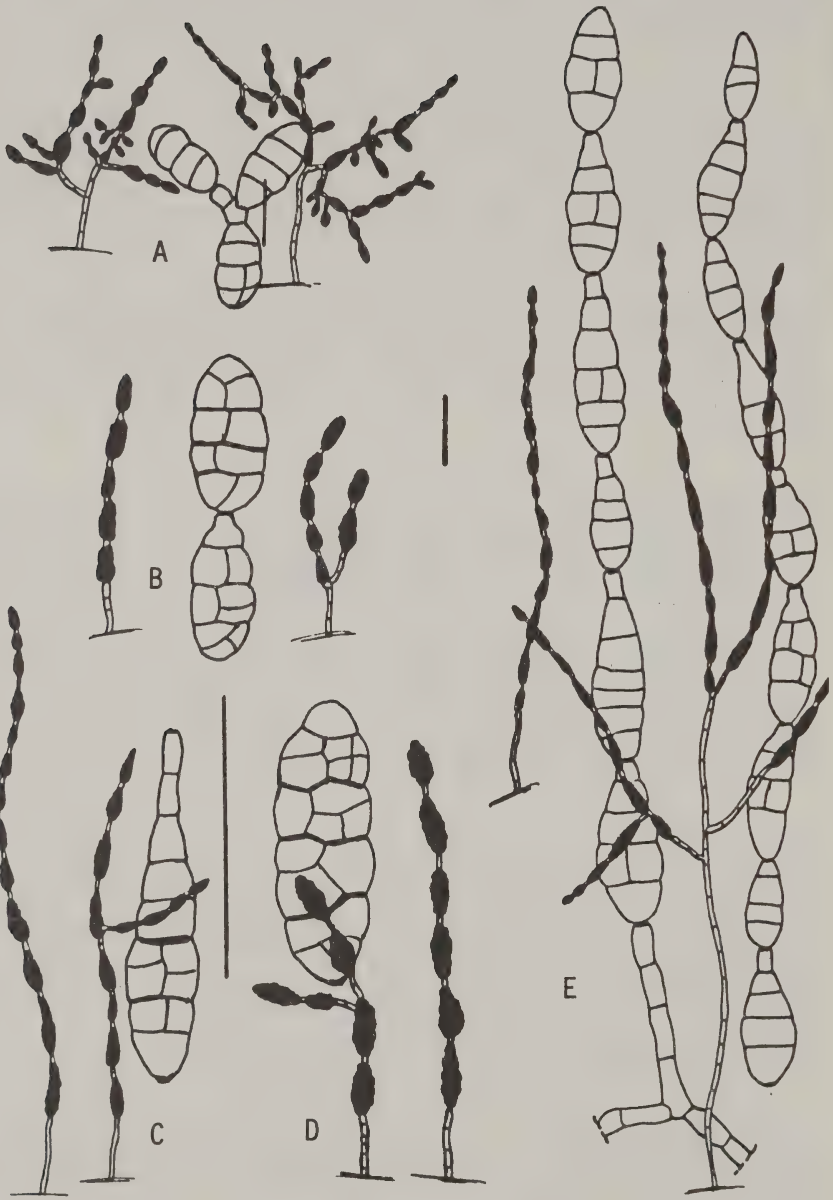

Fig. 173. Sporulation habit and representative or diagnostic conidia: A. *Alternaria alternata*; B. *A. gaisen*; C. *A. tenuissima*; D. *A. toxicogenica*; E. *A. longipes*. Bars = 50μm.

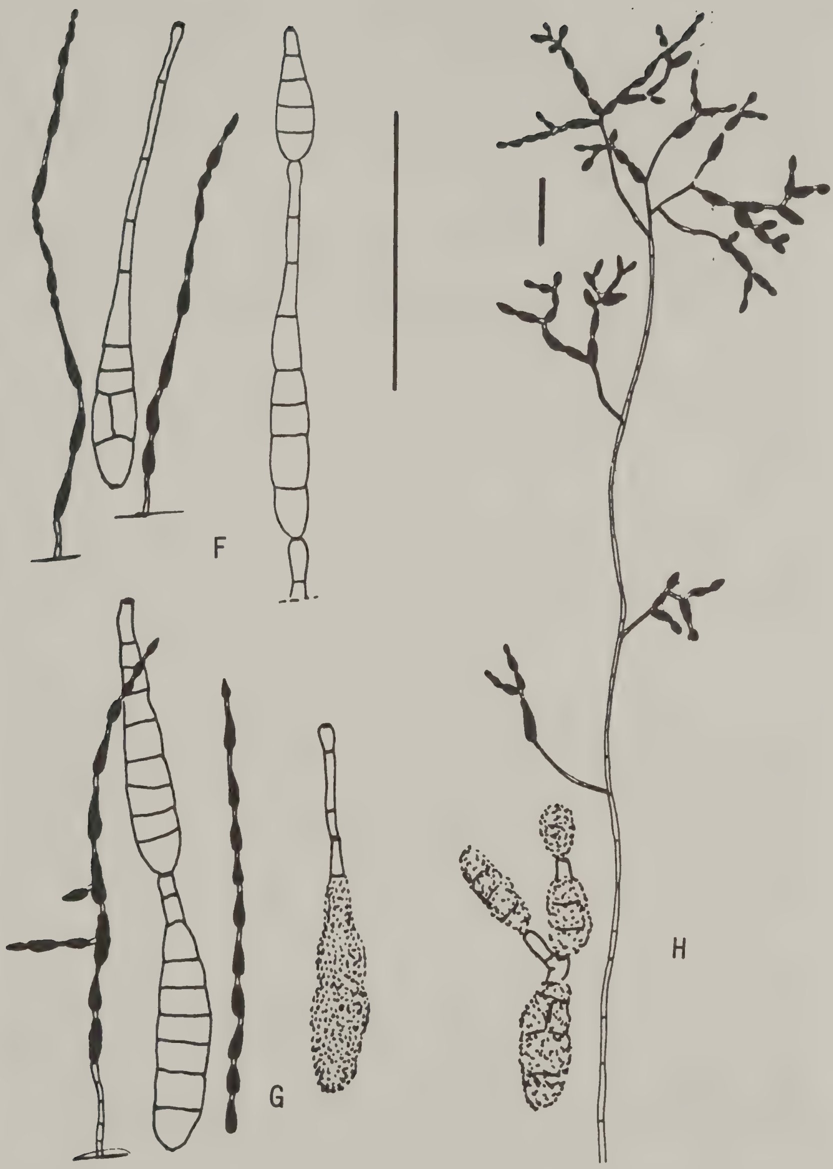

Fig. 173 contd. F. *Alternaria limoniasperae*; G. *A. mali*; H. *A. arborescens*. Bars = 50μm.

Cooke et al. concluded that several isolates of *A. brassicae* (Berk.) Sacc. of diverse geographic origin "showed high levels of [RAPD-PCR based] similarity," which is morphotaxonomically reassuring for this very distinctive pathogen. Their companion conclusion for the 12 other *Alternaria* species also is supportive of current morphology-based opinion to the extent that "little similarity was noted between other species."

Not at all to quibble with such refreshing data, it nevertheless can be noted in HST context that their closest interspecific genetic differences clustered *A. alternata*, *A. citri*, and *A. longipes* (respectively IMI 256370 = EGS 34-039; IMI 082637 = EGS 23-051; IMI 055929 = EGS 07-084). This RAPD-based grouping matches my view that these three species, though individually distinctive, represent a very extensive range of small-spored *Alternaria* taxa at the same "subgenus" level of classification theorized for the ITS results of Kusaba and Tsuge (1995).

But then just one small quibble in discussing this three-taxon interspecific group, which includes supposedly HST-related *A. longipes* and *A. citri*. The authors fall into the trap of associating their IMI-identified *A. citri* isolate with the HST "*A. alternata*...rough lemon pathotype," = "*A. citri*...rough lemon pathotype." The IMI isolate is of a distinctly different species than the "rough lemon pathotypes" of HST studies, these latter being *A. limoniasperae*. The discussion by Cooke et al. of results that show "a close relationship" among these three small-spored species in HST literature neglects a significant point, namely, that the RAPD-focus research and the HST-studies have worked with different isolates and, in some instances, with demonstrably different, sometimes misidentified species, which have been the subject of this current *Alt.T&V* presentation.

* * * * *

My only authoritative stand is that all of the major HST-reported species can be distinguished from each other, and from *Alternaria alternata*, easily and consistently by morphological means—and that to maintain otherwise is simply ignoring demonstrable facts of systematic biology.

The practice in HST research has been to use "*Alternaria alternata* collective species" as a shelter for all of its misidentified toxigenic isolates. However, using the name *A. alternata* necessarily limits the choice—anyone's choice—not to an assemblage of remarkably different species, but to the specific, definable morphologic entity that I have discussed here and elsewhere under the name *A. alternata*. Using this name in other than a one-species sense buries, almost irretrievably, all useful and important information about the morphologically distinct HST-producing species.

The extensive research and the positive import of HST data on plant disease phenomena represent a physiological and biochemical approach to specific pathologies. Pathologists and biochemists, impressed by the undoubted value of this work, have assumed that the conjoined "*A. alternata* collective species" concept has well-considered and predictive classification value. It does not. The HST literature is barren of any body of data or presentation of morphologic insights to support its collective-species concept.

Pathologists with species-oriented interests deserve differential information about the several *Alternaria* species associated with host specific/selective toxicity. Concepts of a nebulous "pathotype" and the equally vague "*Alternaria alternata*" of HST literature do not provide it. Pathologists, and certainly mycotaxonomists, need to reassert the value they place on recognizing distinguishable taxa, named or not, as the operational keys to access the usable accumulations of morphological, pathological and, increasingly, metabolic and molecular information.

Acknowledgements

It is my good fortune to have as reviewers two phytopathologists, Mary E. Palm and Rodney G. Roberts, who also are practicing mycotaxonomists. Both have strong professional interests in *Alternaria* species and their unambiguous relationships to particular pathologies, and thus are able to appraise my arguments and their presentation.

Dr. Palm, working within APHIS/USDA, has been the primary force in arranging my participation in *Alternaria* studies related to diseases of Japanese pear and of citrus cultivars. Her interest and support have increased over the years, as has my gratitude for her efforts.

Dr. Roberts, working within ARS/USDA, has added dimensions of host-susceptibility testing and, increasingly, molecular investigation to our common interest in devising a modern classification scheme for small-spored *Alternaria* species. The influence of his molecular data on my concepts of sporulation groups (they often coincide) is strong and, I hope, will increase.

Individuals and institutions who provided isolates are named in the context of species discussions. Their essential contributions are acknowledged with gratitude.

Wabash College, Crawfordsville, Indiana, continues to support my work through an appointment as Research Associate in the Biology Department faculty, which yields valued access to library and electronic communication facilities.

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MYCOTAXON

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CORTICIOID APHYLLOPHORALES (BASIDIOMYCOTA) FROM THE PATAGONIAN ANDES FORESTS OF ARGENTINA
4. NOTHOCORTICIUM PATAGONICUM GEN. ET SP. NOV.

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Abstract

Nothocorticium patagonicum is described as a new genus and species. It is characterized by effused basidiocarps that develop marginal hyphal strands, smooth hymenophore, large, two-spored basidia with long sterigmata, large, cylindric, thin-walled spores that are white in print and encrusted, thick-walled, generally branched hyphidia. It is found on bark of living trees and fallen wood of *Nothofagus* spp. and belongs in the Vuilleminiaceae, it also seems allied to the genus *Corticium*, especially *C. quercicola*, from which it differs in the lack of probasidia and the presence of euhymenium. Its relationship to *Corticium, Dendrocorticium* and *Vuillieminia* is discussed.

Key words: Aphyllophorales, Corticiaceae, *Nothocorticium*, *Nothofagus*.

Introduction

A peculiar corticioid fungus was found recently on bark of living trees and fallen wood of *Nothofagus pumilio* (Poepp. & Endl.) Krass. and *Nothofagus dombeyi* (Mirb.) Oerst. in southern South America. This taxon is described herein in a new genus *Nothocorticium*.

Materials and Methods

Most specimens are kept at the authors' laboratory but the type and/or duplicate specimens have been deposited at BAFC and DAOM (Holmgren *et al.*, 1990). Spores were measured in 5 % KOH aqueous plus 1% phloxine and statistical calculations were made on measurements from rehydrated material; N= number of measured spores, L= mean length, and W= mean width. Microscopic sections of basidiocarps were examined in Melzer's Reagent (Hawksworth *et al.*, 1995) for amyloid or dextrinoid reactions, and Cotton Blue in lactic acid (Hjortstam *et al.*, 1987) for cyanophily. Common color names have been used along with color notations according to Munsell (1990). AG and MR refer to collections made by the first and the second author.

Results

Nothocorticium patagonicum Greslebin & Rajchenb. gen. et sp. nov. Fig. 1-4

Fructificatio resupinata, effusa vel leviter reflexa, velutina, persicina vel luteo aurea. Subiculo gossypino, cremeo, 120-250 µm crasso. Hyphae generatoriae afibulatae. Basidia clavata elongatis, 2-sterigmaticus, 60-140 x 10-18 µm. Basidiosporae cylindraceae vel leviter curvatae, (20-)25-42 x (7-)8-10 µm. Dendrophyses nullus. Hyphidia perabundantibus, ramosae, crassetunicatae, incrustatae. Holotypus BAFC 34525.

Basidiocarp resupinate, effused, thinning out towards margin, forming small orbicular or oblong patches up to 7-30 x 5-14 mm, that may coalesce up to 60 x 40 mm, 120-250 µm thick. Hymenial surface smooth, subceraceous when fresh, velutinate, felty or loosely felty under the lens, peach-colored (very pale brown 10YR 8/3-8/4, pink 7.5YR 8/4, or pink reddish yellow 7.5YR 7/4-7/6) to light yellow or light mustard in color. Margin effused, sometimes raised, fibrillose or mycelial, white or concolorous with the hymenial surface, forming white and branched hyphal strands that develop profusely into the bark. Subiculum cottony and white, contrasting with the hymenial surface.

Hyphal system monomitic, generative hyphae simple septate, 2-5 µm in diam., with hyaline, thin to thickened walls. A basal layer of horizontally arranged, thick-walled hyphae is formed against the substrate. Hyphal strands up to 150 µm diam, composed of parallel hyphae, 2-4(-6) µm diam, thin- and thick-walled, with simple septa, some of them ampullaceous; hyphae of the surface shows the same encrustation than the hyphidia.

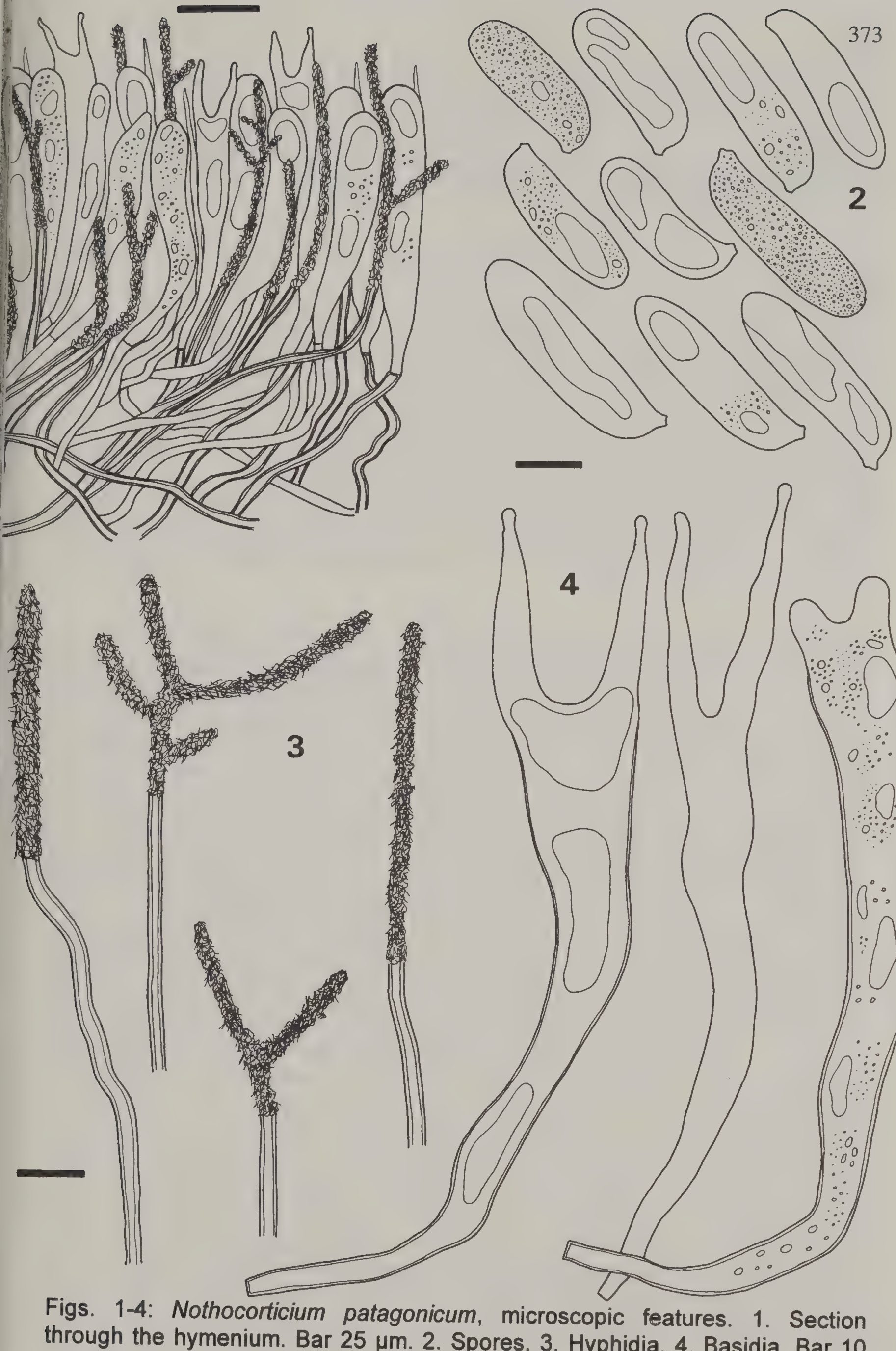
Hymenium euhymenium composed of basidia, basidioles and hyphidia.

Basidia narrowly clavate, sinuous, 60-140 x 10-18 µm, with two prominent sterigmata up to 35 µm long, simple septate at the base. In some specimens very difficult to find because they turn hyaline, empty and finally collapse.

Basidioles: subulate to clavate, 40-100 x 6-9 µm, filled with oil drops.

Hyphidia abundant throughout the fruitbody, originating from generative hyphae and projecting up to 20(-30) µm, IKI-, acyanophilous; of two types: thick-walled, strongly encrusted, (3-)4-5(-7) µm diam, most simple but some with straight branches; and filiform, 2-4 µm diam, thin-walled, lacking encrustation, scarce.

Basidiospores cylindrical, some slightly curved, variable in size, 20-27(-35) x 7-9 µm from specimens on *Nothofagus pumilio*, or (20-)25-42 x (7-)8-10 µm from specimens on *N. dombeyi*, hyaline, thin-walled, inamyloid, guttulate and with a distinct, blunt apiculus. The point of attachment to the sterigma is not the apiculus itself but to one side of it.



Figs. 1-4: *Nothocorticium patagonicum*, microscopic features. 1. Section through the hymenium. Bar 25 µm. 2. Spores. 3. Hyphidia. 4. Basidia. Bar 10 µm.

Spore measurements:

Specimens	N	L ± σ (μm)	W ± σ (μm)
Holotype on *N. dombeyi*	23	33.60 ± 6.42	9.04 ± 0.87
AG 51 and 257 on *N. pumilio*	18	25.27 ± 3.15	8.14 ± 0.70

Habitat: On bark of living *Nothofagus dombeyi* and *N. pumilio* and on fallen wood of *N. pumilio* (Fagaceae).

Material studied: On *N. pumilio*: ARGENTINA, TIERRA DEL FUEGO, Estancia El Valdez, leg. AG 51 (BAFC 34526), 29.Nov.95. Ibid., leg. ipse AG 359 and 360, 5.Mar.96. Ibid., leg. ipse AG 412, 5.Mar.96. Ibid., leg. ipse AG 436, 5.Mar.96. Ibid., Río Valdez, leg. ipse AG 257, 4.Mar.96. ARGENTINA, CHUBUT, Río Senguer, Lago La Plata, leg. AG and MR 531, 27.Mar.96. Ibid., leg. ipse AG 542 and 543, 28.Mar.96. Chubut, Los Alerces National Park, Cerro Dedal, leg. AG and MR 1058, 9.May.97. On *N. dombeyi*: ARGENTINA, CHUBUT, Los Alerces National Park, Río Arrayanes, camping area, leg. MR and AG 11270-11274, 19.Jun.97. Chubut, Los Alerces National Park, arroyo Coliguay ca. Lago Rivadavia, leg. MR and AG 11275 (BAFC 34525, Holotype) and 11276, 19.Jun.97. ARGENTINA, RÍO NEGRO, Bariloche, Mallín Ahogado, Cajón del Río Azul, leg. AG 1649, 15.May.98.

Remarks: *Nothocorticium patagonicum* is characterized by large 2-sterigmate basidia, large basidiospores with a blunt apiculus and predominately thick-walled, narrow generative hyphae that culminate in the hymenium as simple or branched, strongly encrusted hyphidia. The specimens on *Nothofagus dombeyi*, including the type, were mature and displayed basidia and relatively abundant basidiospores of the larger size. On the contrary, the specimens gathered on *N. pumilio* seemed to be overmature, with few or no basidia and few basidiospores, the latter of smaller size. As indicated above the spore sizes overlap, but basidiospores from specimens gathered on *N. pumilio* do not reach the larger sizes. Because the general macro and microscopical features are identical in all specimens we assume that all belong to the same taxon.

By the color of the basidiocarp and the presence of hyphidia, *Nothocorticium patagonicum* resembles the genus *Corticium* Pers., especially *Corticium quercicola* Jülich (= *Laeticorticium quercinum* J. Erikss. & Ryv.), which also has very large basidia with two prominent sterigmata and simple septate hyphae. However, *C. quercicola* has probasidial bladders characteristic of the genus *Corticium*, while *Nothocorticium* lacks them. Furthermore, *Corticium* sensu stricto gives pink spore print (Boidin & Lanquetin, 1983) and possesses a catahymenium, while *Nothocorticium* gives a white spore print and forms an euhymenium. *Dendrocorticium* M.J. Larsen & Gilb., though presenting a white spore print (Boidin & Lanquetin, 1983), also differs in having probasidial bladders (Larsen & Gilbertson, 1974). The inclusion of this species in the genus *Vuilleminia* Maire was considered on the basis of basidial and basidiospores' shape and size, but this possibility is rejected because the species lacks a gelatinous texture, a habit under peeling bark and dendrohyphidia (Boidin *et al.*, 1994). It was not included in *Dendrothele* v. Höhn. & Litsch., in spite of sharing a similar habit, large and oily basidia, and

large basidiospores, since the species lacks the typical cretaceous texture of *Dendrothele*, the catahymenium and the typical dendrophyses (but *cfr.* Greslebin & Rajchenberg, 1998).
The posibility of including this taxon in *Clavulicium* Boid. was also evaluated, but it differs in lacking hyphidia and in having smaller basidiospores and basidia.

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CULTURAL STUDIES OF AGROCYBE CYLINDRICA, GYMNOPILUS PAMPEANUS AND OUDEMANSIELLA CANARII (AGARICALES) ISOLATED FROM URBAN TREES

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SUMMARY

Cultural studies of three species of Agaricales isolated from trees, *Agrocybe cylindrica*, *Gymnopilus pampeanus* and *Oudemansiella canarii*, were undertaken. Although these are well-known species, a complete cultural study of them has never been made. Nobles' code is given for each.

INTRODUCTION

During the last fifty years, the use of cultural characters in taxonomy has increased, mostly concerning the wood-rotting Aphyllophorales (Davidson et al, 1942; Nobles 1948, 1958, 1965; Stalpers, 1978, Nakasone, 1990; Wright and Deschamps, 1972, 1974). At this time, there are few published reports of cultural studies of Agaricales in which formal descriptions have been made (Campbell and Petersen, 1975; Thorn and Barron, 1986; Ovrebo, 1988; Vilgalys and Miller, 1983; Mueller, 1984; Semerdzieva, 1965a, 1965b; Pantidou 1962; Pantidou and Groves, 1966; Pantidou and Watling, 1973; Pantidou et al, 1983; Jacques-Felix, 1968; Desjardin, 1990; Petersen, 1992; Petersen and Krisai-Greilhuber, 1996).

In a survey of the xylophilous mycota present in the urban parks of Buenos Aires, Argentina, cultures of three species of Agaricales, *Agrocybe cylindrica* (DC: Fr.) Maire, *Gymnopilus pampeanus* (Speg.) Singer and *Oudemansiella canarii* (Jungh.) Hoehnel, growing on *Acer negundo* L., *Eucalyptus viminalis* Labill. and *Erythrina crista-galli* L. respectively, were isolated from fruitbodies. The aim of this study was to describe these three species of Agaricales in culture.

MATERIALS AND METHODS

Pure cultures were isolated by inoculating tube slants with hyphae aseptically obtained from fresh tissue of the fruitbodies. The cultural studies of the three species and culture codes both follow the system of Nobles (1965).

Cultures were grown on malt-extract agar in the dark at 25 °C. Growth rate was expressed as the number of weeks required by the cultures to cover the surface of a nine cm Petri dish from an inoculum placed at the side of the dish.

Oxidase reactions were recorded one week after incubation in the dark at 25 °C. Gallic acid (GAA) and tannic acid (TAA) agar media were made according to Nobles (1965); tyrosine, paracresol and guaiacol 0.2% agar media were made according to Boidin (1954). The relative intensity of the reaction is indicated with plus or minus signs.

The abbreviation BAFC stands for the culture collection of the Department of Biological Sciences, School of Exact and Natural Sciences, University of Buenos Aires. Fruit bodies were annotated, dried, and deposited in the mycological herbarium (BAFC).

RESULTS AND DISCUSSION

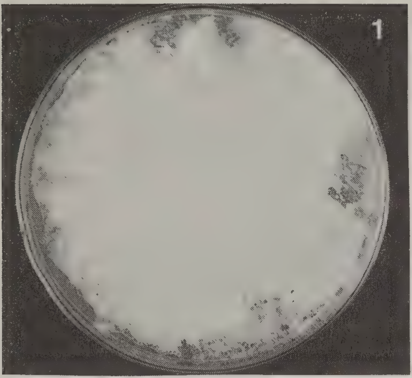
Agrocybe cylindrica (DC: Fr.) Maire, Mem. Soc. Scie. Nat. du Maroc 45: 106. 1938

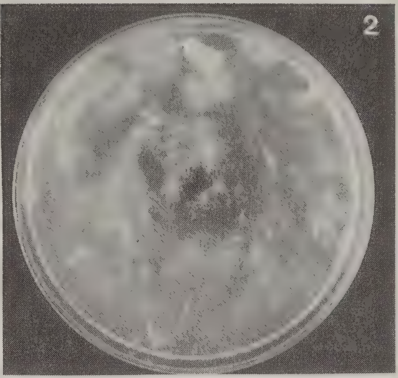
Macroscopic characters: growth moderate, dishes covered at the 3rd week. Margin even. Mat homogeneously silky, white. Dark brown zones develop around the inoculum and scatter on the agar towards the end of the study (Fig. 1). Odor none. Reverse brown in patches corresponding to the colored mycelium.
Oxidase reactions: gallic acid -, without growth; tannic acid -, without growth ; tyrosine ++ (yellowish-orange reaction), with growth; guaiacol -, without growth; paracresol -, without growth.
Microscopic characters: Advancing zone and aerial mycelium: generative hyphae clamped, staining in phloxine, hyaline, regularly branched, 1,5(-2)-4(-5) µm diam (Fig. 5, A-B), occasionally diverticulate (Fig. 5, C); chlamydospores terminal or intercalary, elliptic to globose and irregularly shaped, 5-12 x 6-20 µm, with slightly thickened walls, containing many guttate vesicles (Fig. 5, D), produced from subtending thinner hyphae, 1,5-2 µm diam. Hyphae with numerous irregular thick-walled branches, developing a plectenchyma or textura angularis *sensu* Desjardin (1990). (Fig. 5, E-F), imparting a dark brown color to the mat.
Species code: 1.3.11.34.37.39.43.54
Material studied: Argentina, Capital Federal, BAFC 2217, 4.IV.94; BAFC 2219, 14. IV.94; BAFC 2220, 28.IV.94; BAFC 2223, 14.IV.94; BAFC 2227, IV.94. All isolates from fruitbodies on standing *Acer negundo*.

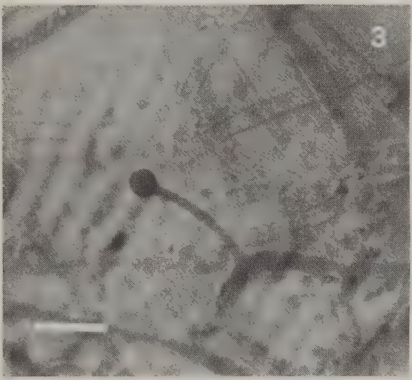
During this study, *Agrocybe cylindrica* was only found on *Acer negundo*. It has been recorded in Argentina on the following hosts: *Melia azedarach*, *Populus alba*, *P. angulata*, *Robinia pseudoacacia*, *Brussonettia papyrifera*, *Quercus* sp., *Acer negundo*, *Salix* sp., *Allophylus edulis*, *Cupania* sp., *Phoebe* sp., and *Maytenus* sp. (Singer, 1950; Singer, 1969 and Singer & Digilio, 1952). As the last four ones are native hosts, we assume that this species has escaped to them.

Gymnopilus pampeanus (Speg.) Singer, Lilloa 22: 561. 1951

Note: Singer gives (1975, 1986) this citation but he also says that the taxon may be considered as a subspecies of *G. spectabilis*.
Macroscopic characters: growth moderate, plates covered at the third week. Mat downy, becoming cottony, more or less lacunose, with plumose outgrowths, white (Fig. 2). Advancing zone submerged. Margin uneven. Reverse unchanged.
Oxidase reactions: gallic acid ++++, without growth; tannic acid ++++, without growth; tyrosine ++, with growth; guaiacol -, without growth; paracresol +++, without growth.







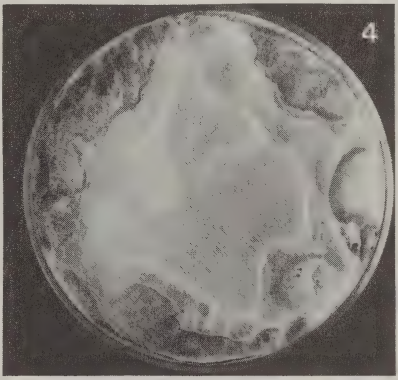


Fig. 1 *Agrocybe cylindrica*. Mat at 6th week.
Fig. 2, 3 *Gymnopilus pampeanus*. 2: mat at 6th week. 3: capitate apice of narrow, tapering hyphal branch. Scale line: 10 µm.
Fig. 4 *Oudemansiella canarii*. Mat at 6th week.

Microscopic characters: Advancing zone: generative hyphae clamped, staining in phloxine, hyaline, branched, 2-3 µm diam (Fig. 6, A-B). Aerial mycelium: hyphae similar to those in the advancing zone with numerous clamp connections, very frequently branched; chlamydospores terminal or intercalary, elliptic to globose, 9-18 x 6-9 µm (Fig. 6, C). Other noteworthy characters: transversal anastomoses, some with clamps (Fig. 6, D). Capitate apices, 3-5 µm diam, of narrow, tapering hyphal branches. (Fig. 3 and 6, E).
Species code: 2.3.7.34.36.38.44.54.58
Material studied: Argentina, Capital Federal, BAFC 2200, isolated from fruitbody on standing *Eucalyptus viminalis*, 6.V 94.

G. pampeanus always occurred during this study on *Eucalyptus* sp., and there is no evidence it has escaped to native trees. We agree with Singer (1950), who always

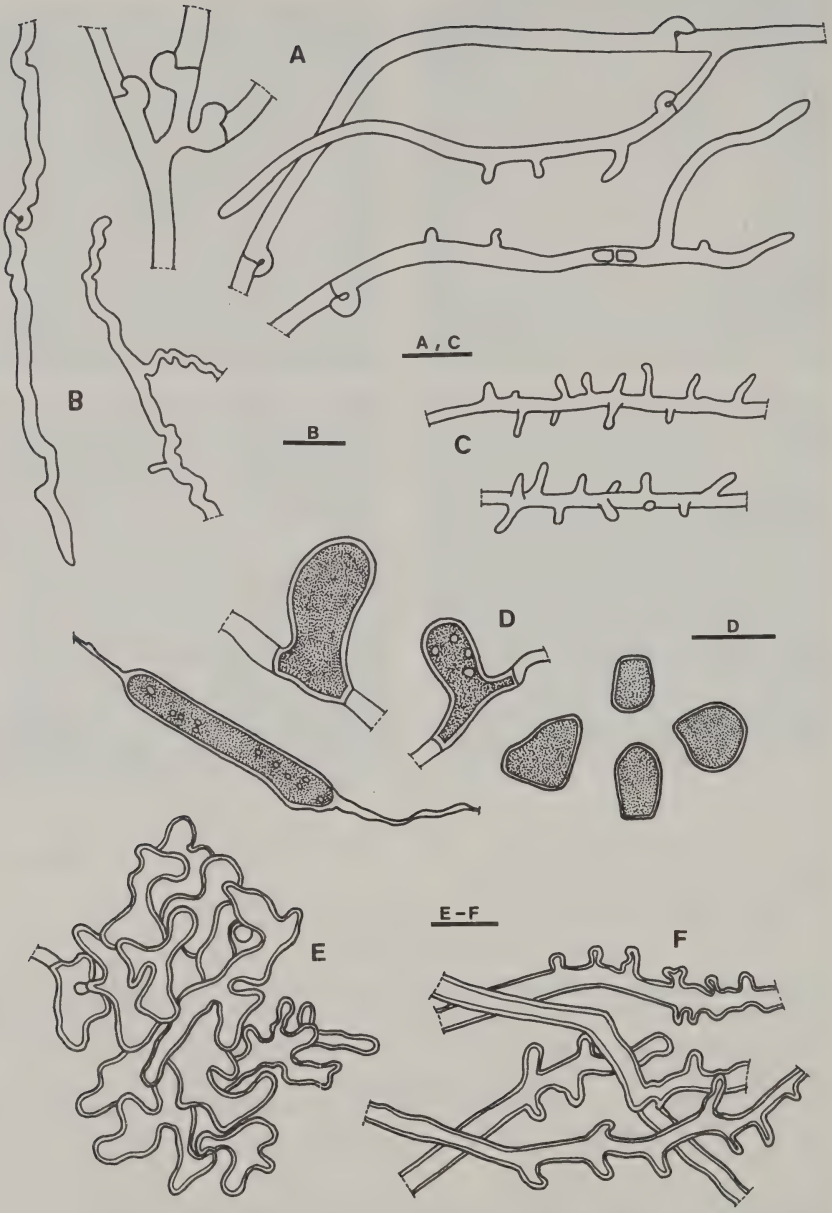


Fig. 5 (A - F) *Agrocybe cylindrica*. A-B: generative hyphae. C: diverticulate hyphae. D: chlamydospores. E: hyphae with irregular thick walled branches. F: plectenchyma. Scale line: 10 µm.

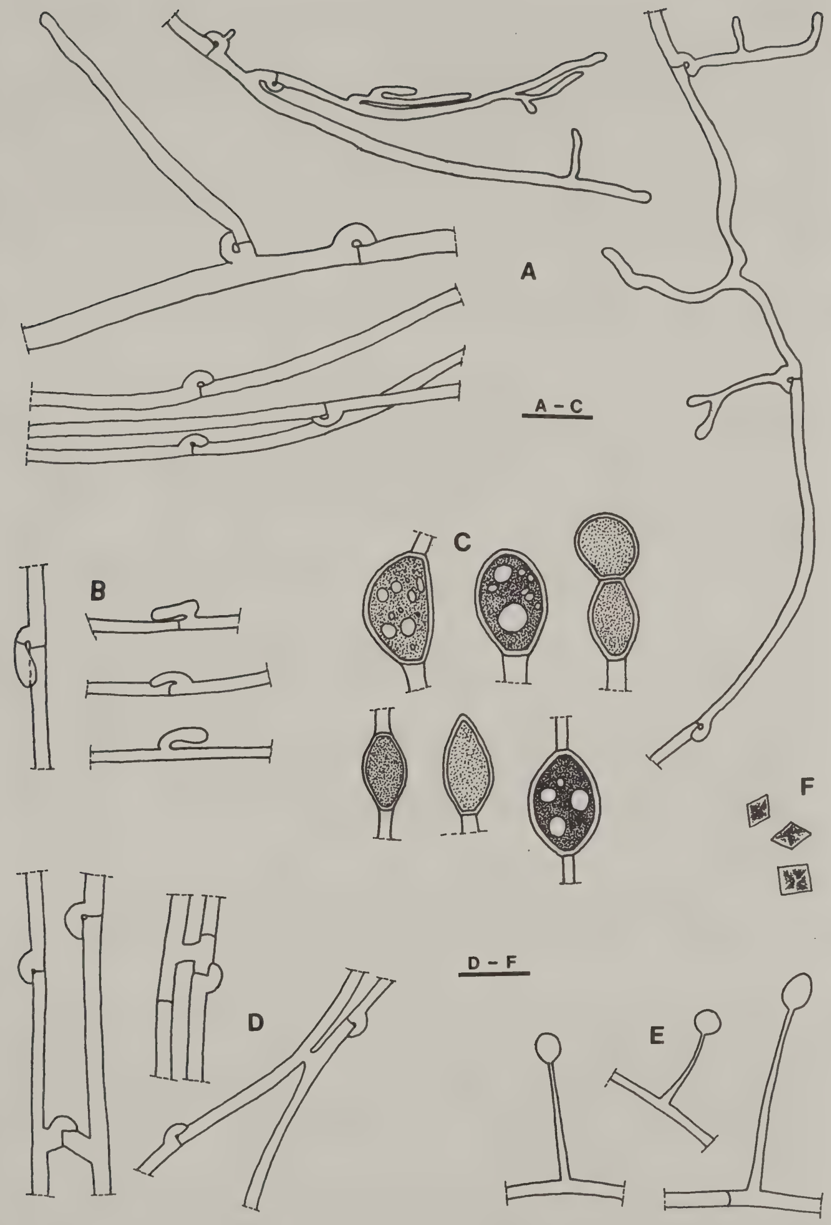


Fig. 6 (A - F) *Gymnopilus pampeanus*. A: generative hyphae. B: hyphae with short branches that seem to be clamps. C: chlamydospores. D: anastomoses. E: capitate apices of narrow, tapering hyphal branches. F: crystals. Scale line: 10 µm.

found it in wounds on trunks of living trees, particularly at the bases of *Eucalyptus* sp. and rarely on other trees.

Capitate apices of narrow, tapering hyphal branches found in *Gymnopilus* cultures seem to be at first sight the nematode-trapping droplets that have been observed in several species of the genus *Pleurotus* (Hibbett and Thorn, 1994; Thorn and Barron, 1984; Thorn and Tsuneda, 1993; Petersen and Krisai-Greilhuber, 1996) and that have been accepted as a generic character (Hibbett and Thorn, 1994). As droplets over these structures were not observed, we could not assume they were nematode-trapping structures. It would be of great interest to study in culture other species of the genus to know if this particular structure is a generic character.

Oudemansiella canarii (Jungh.) Hoehnel. Akad. Wiss. Wien, Math-naturw. Kl. 118: 276. 1909

Macroscopic characters: growth moderate, plates covered at the third week. Mat woolly, then becoming cottony, white. Dark brown zones scattered on the agar from the second week and up to the end of the study (Fig. 4, A). At the fourth week, primordia are formed on the brown dark zones. Not fruiting by sixth week. Advancing zone submerged. Margin even. Odor none. Reverse brown in parts corresponding to the colored mycelium.
Oxidase reactions: gallic acid +++++, without growth; tannic acid +++++, without growth; tyrosine -, with growth; guaiacol +++, without growth; paracresol -, without growth.
Microscopic characters: Advancing zone: generative hyphae branched, with few clamp connections, (1,5-)2-4 µm diam. Aerial mycelium: generative hyphae with numerous clamp connections, branched, (1-)1,5-6(-7) µm diam (Fig. 7, A). Arthrospores 2,5-3(-4) x 5-11 µm (Fig. 7, B). Agar-surface hyphae with numerous irregular thick-walled branches, composing a plectenchyma or textura angularis and epidermoidea sensu Desjardin (1990), imparting a dark brown color to the mat. (Fig. 7, E-F).
Other noteworthy characters: 1) transversal anastomoses (Fig. 7, D); 2) a few intercalary rings (Fig. 7, C).
Species code: 2.3.11.35.37.38.43.54.
Sexuality: Amphithallic (Petersen and Halling, 1993)
Material studied: Argentina, Capital Federal, BAFC 2194, isolated from fruitbodies on dead *Erythrina crista-galli*, 14.IV.94.; BAFC 2176, III. 95.

We have compared published cultural studies of different species in the Xerulaceae with that of *O. canarii*. Production of brown crustose patches or zones composed by a textura epidermoidea is shared by several species of *Xeromphalina* (Redhead, 1987; Nobles 1965; Petersen 1991), *Xerula*, including *X. furfuracea* and *X. megalospora* (Redhead et al, 1987) and *Oudemansiella mucida* (in single spore isolates) (Petersen and Halling, 1993).

Tests for extracellular oxidase in *O. canarii* were positive for laccase (gallic acid agar, tannic acid agar and guaiacol) and negative for tyrosinase (tyrosine and p-cresol). These results are consistent with those for *Xeromphalina campanella* and *Xeromphalina*

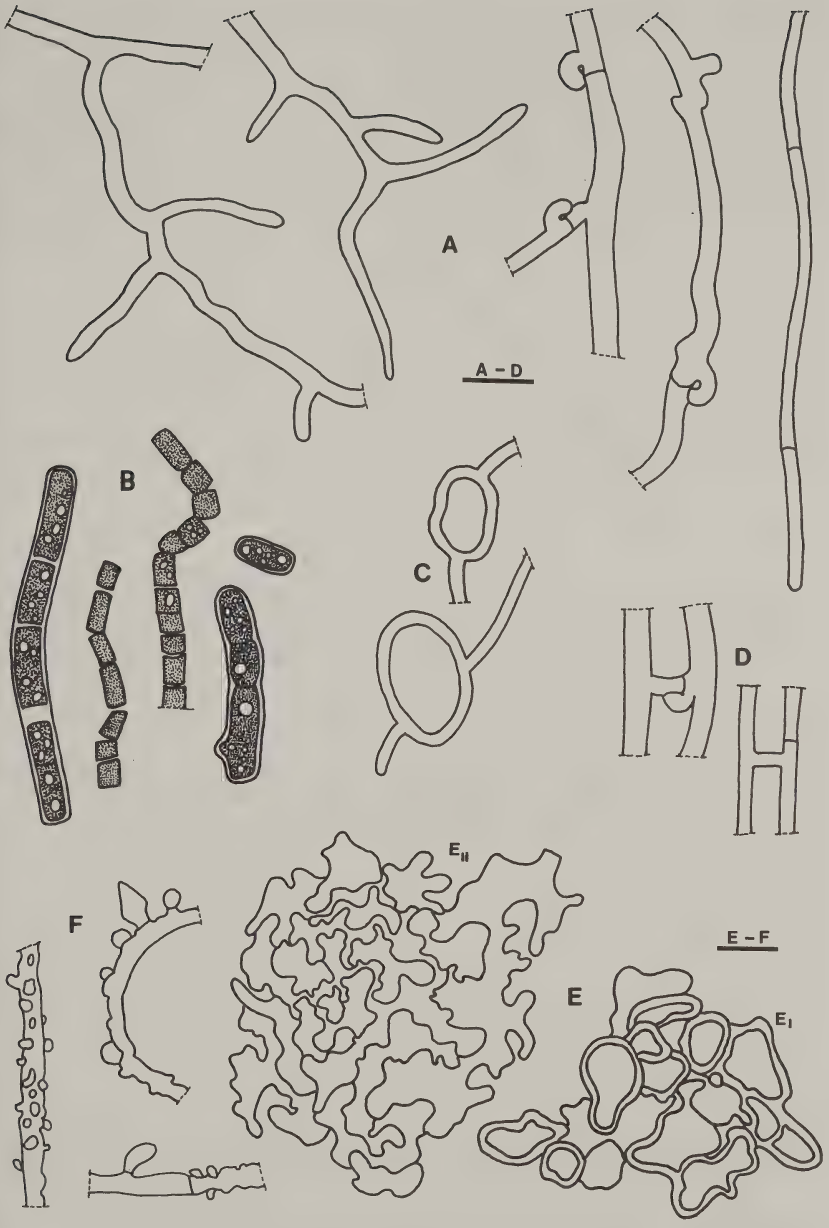


Fig. 7 (A - F) *Oudemansiella canarii*. A: generative hyphae. B: Arthrospores. C: intercalary rings. D: anasthomosis. E: plectenchyma. F: hyphae with irregular protuberances. Scale line: 10 µm.

kauffmanii, but not for *X. brunneola* which lacks laccase production (Petersen, 1991). *Xerula furfuracea* and *X. megalospora* are laccase positive and tyrosinase negative, although there is some variation between isolates (Redhead et al, 1987).

Nobles (as *Collybia velutipes*, 1965) describes the presence of arthrospores in *Flammulina velutipes*. This character has not been found in the cultures of *Xerula* and *Xeromphalina* species. The production of asexual propagules appears to be absent in *Oudemansiella mucida* and *O. brunneomarginata* and according to Petersen and Halling (1993), *O. canarii* only produced them after growth on oxgall agar.

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STUDIES IN CHAETOSPHAERIA SENSU LATO I. THE GENERA CHAETOSPHAERELLA AND TENGIOMYCES GEN. NOV. OF THE HELMINTHOSPHAERIACEAE

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ABSTRACT

Species currently placed in *Chaetosphaeria* sensu lato that have the character of transversely septate, versicoloured ascospores in common are discussed in a series of publications. Revision of their type material revealed that they are not congeneric with the type species of *Chaetosphaeria* and with each other. The families Chaetosphaeriaceae, Helminthosphaeriaceae, Lasiosphaeriaceae and Trichosphaeriaceae are discussed. The present paper deals with species of the genus *Chaetosphaerella* with *Oedemium* and *Veramycina* synanamorphs, including material in the Czech Republic, China, Brazil, France, India, Italy, Sri Lanka and U.S.A. Two species are accepted in *Chaetosphaerella*, viz. *C. fusca* and *C. phaeostroma*. They are described and illustrated and listed along with their full synonymy and notes on previous descriptions and illustrations. A key to the species is provided. *Chaetosphaerella fusispora* is excluded from the genus, its relationships with *Chaetosphaerella* and other genera are discussed. The new genus *Tengiomyces* is described for the single species *Tengiomyces indicus* (syn. *Chaetosphaerella indica*), a lignicolous, saprobic fungus. An associated hyphomycete, *Spadicoides* sp., is presumed to be the anamorph. Additional species *Chaetosphaeria bihyalina*, *C. incrustans*, *C. iquitosensis* and *C. phaeostromoides* are considered and their relationships with *Chaetosphaerella* are disscused. On the basis of the morphological characters of perithecium, ascus and conidial development *Chaetosphaerella* and *Tengiomyces* are assigned to the Helminthosphaeriaceae.

Key words. Ascomycotina, wood-inhabiting fungi, systematics.

INTRODUCTION

In this series of publications, species that share the characters of small, superficial, nonstromatic, black, setose or glabrous perithecia and septate, versicoloured ascospores are reviewed and discussed. They are currently placed in diverse genera of pyrenomycetous ascomycetes, mostly in *Chaetosphaeria* Tul. & C. Tul. and less often in *Chaetosphaerella* E. Müll. & C. Booth and *Lasiosphaeria* Ces. & de Not. of the Lasiosphaeriaceae Nannf. (Barr, 1990). Versicoloured ascospores with the central cells becoming brown and the terminal cells remaining hyaline are characteristic of twenty-eight species. Examination of their type and other herbarium material showed that those species differ radically from the generic concepts of *Chaetosphaeria*, *Chaetosphaerella* and *Lasiosphaeria*. These species need to be transferred to modern genera and should be classified as members of the Chaetosphaeriaceae Réblová, M.E. Barr & Samuels (Sordariales Chadef. ex D. Hawksw. & O.E. Erikss.) and the Helminthosphaeriaceae Samuels, Candoussau & Magni and Trichosphaeriaceae G. Winter (Trichosphaeriales M.E. Barr). The affinities of these species lie rather with these families than with the Lasiosphaeriaceae Nannf. (Sordariales) where they were previously included (Barr, 1990).

The Lasiosphaeriaceae Nannf. sensu Lundqvist (1972), Barr (1990) and Eriksson & Hawksworth (1993) is a broadly perceived family of the Sordariales and does not appear to be monophyletic. The Lasiosphaeriaceae was recently narrowly limited by Réblová & al. (1998) for core genera of the family such as *Apiosordaria* Arx & W. Gams, *Lasiosphaeria*, *Bombardia* (Fr.) P. Karst., *Cercophora* Fuckel and *Eosphaeria* Höhnel that form a homogenous group characterized by features of peridium that is leathery, partly carbonaceous in outer layers and lightly pigmented brown, comprises angular to brick-like cells, perithecial apex not differentiated from the rest of the perithecium; presence of subapical chamber and plasmatic globule in the ascal apex; persistent, narrowly cylindrical, paraphyses and periphyses; mostly cylindrical ascospores, often bent in the lower third, becoming often inflated and dark pigmented in the upper part, germinating by both phialide and germ tube; anamorphs exclusively with enteroblastic-phialidic conidiogenesis (Gams & Holubová-Jechová, 1976; Mouchacca & Gams, 1993; Petrini & al., 1984). Species are lignicolous, herbicolous, fimicolous and rarely terricolous.

The Chaetosphaeriaceae forms a homogenous unit of genera such as *Ascocodinaea* Samuels, Candoussau & Magni, *Chaetosphaeria* Tul. & C. Tul., *Melanochaeta* E. Müll., Harr & Sulmont, *Melanopsammella* Höhnel, *Porosphaerella* E. Müll. & Samuels, *Porosphaerellopsis* Samuels & E. Müll. and *Striatosphaeria* Samuels & E. Müll. Réblová & al. (1998) separated

Chaetosphaeriaceae from the Lasiosphaeriaceae on the basis of different characters of peridium anatomy (thin, brittle peridium of thin-walled, opaque brick-like cells), ascal (lack of subapical chamber and plasmatic globule) and ascospore (ellipsoidal to fusiform, 1-3-septate, germinating by germ tube) anatomy and a homogenous group of anamorphs, all of which are dematiaceous hyphomycetes that have exclusively enteroblastic-phialidic conidiogenesis. Species are lignicolous or fungicolous.

The characters of holoblastic conidiogenesis and features of perithecial, ascal and ascospore anatomy were suggested by Réblová & al. (1998) in distinguishing Helminthosphaeriaceae and Trichosphaeriaceae from the Lasiosphaeriaceae and Chaetosphaeriaceae. The two former families placed in the Trichosphaeriales (Barr, 1990; Réblová & al., 1998) proved to be a home for several other species examined. The Helminthosphaeriaceae (Samuels & al., 1997) with the single genus *Helminthosphaeria* Fuckel emend Samuels, Candoussau & Magni accommodates pyrenomycetous ascomycetes with nonstromatic, setose or glabrous, rarely collapsing perithecia; brittle, wide, peridium that comprises melanized, thick-walled, pseudoparenchymatous cells forming low warts, perithecial apex truncate, often formed of conspicuously diverging, more or less clavate hyphal elements; setae arise as outgrowth of the hyphal elements as well as form from the entire surface of the perithecium; ostiolar canal periphysate; cylindrical paraphyses; apical ring J-, simple or lacking; ascospores ellipsoidal, 0-several-septate, sometimes with a pore at both ends, germinating by germ tube; anamorphs with holoblastic-tretic conidiogenesis. Species are fungicolous. The peridium, ascus and ascospore anatomy of the Helminthosphaeriaceae is illustrated for *Helminthosphaeria* by Samuels & al. (1997).

Although the Trichosphaeriaceae superficially resembles the Chaetosphaeriaceae in having small, black, setose or glabrous perithecia, it is distinct because of its leathery to firm, only partly carbonaceous peridium that comprises lightly coloured, angular to brick-like cells; periphyses arising from the inner meristem of thin-walled cells in the perithecial apex that is distinct from a meristem from which the paraphyses arise; ascospores with 0-1 to several transverse septa or dictyoseptate; conidiogenesis holoblastic. Species are lignicolous. In the Chaetosphaeriaceae periphyses arise from the same meristem as paraphyses.

A brief note on the telemorph-anamorph connections of these families follows. The anamorphs of the Trichosphariaceae are poorly known, e.g. *Rhamphoria* (Müller & Samuels, 1982). In the Chaetosphaeriaceae, Lasiosphaeriaceae and Helminthosphaeriaceae, where the anamorphs are better known, it is not unusual for more than one teleomorph genus to have an anamorph genus in common. This apparently indicates that those genera that have common anamorphs could be closely related. We must also consider the possibility that the anamorph genera might be artificial or maybe the teleomorph genera are not actually separable. What may be more significant is how the conidia form and also the conidial morphology. On one hand, genera that are associated with the same anamorph genus are clearly distinct in peridium anatomy and hamathecium and ascal and ascospore anatomy. For instance *Cercophora, Chaetosphaeria, Bombardia, Eosphaeria, Lasiosphaeria* have *Phialophora* anamorphs or synanamorphs; *Chaetosphaeria*, *Ascocodinaea* and *Striatosphaeria* have *Dictyochaeta* Speg. anamorphs; *Chaetosphaeria* and *Melanochaeta* have *Chalara* (Corda) Rabenh. anamorphs or synanamorphs; *Lasiosphaeria* and *Iodosphaeria* Samuels, E. Müll. & O. Petrini, have *Selenosporella* Arnaud ex Macgarvie anamorphs or synanamorphs and *Phaeotrichosphaeria* and *Lasiosphaeria* have *Endophragmiella* anamorphs or synanamorphs. On the other hand, the association with anamorph genera that are characteristic by rather simple conidiophore anatomy and conidiogenous cells would not seem to be highly significant for establishing close relationships among morphologically distinct teleomorphs. For example the anatomically simple genus *Phialophora* is known to be an anamorph of different groups of Ascomycetes, e.g. Chaetosphaeriaceae (Constantinescu & al., 1995; Réblová, 1998a), Chaetomiaceae G. Winter (Udagava & Muroi, 1981), Dermateaceae Fr. (Cain, 1952; Kendrick & Carmichael, 1973), Lasiosphaeriaceae (Gams & Holubová-Jechová, 1976; Gams, 1973) Herpotrichiellaceae Munk (Schol-Swarz, 1968) and Magnaporthaceae P. Cannon (Cannon, 1994).

The majority of species discussed in this series of publications was currently placed in *Chaetosphaeria* and only few in *Chaetosphaerella* and *Lasiosphaeria*. The detailed description of *Chaetosphaeria* and an account of accepted species are provided by Réblová & al. (1998). A short historical resumé of the genus follows. *Chaetosphaeria* was introduced by Tulasne & Tulasne (1863) and later re-established by Booth (1957, 1958) for nonstromatic pyrenomycetous ascomycetes with superficial, globose to subglobose perithecia with a brittle perithecial wall that comprises thin-walled, opaque, dark brown brick-like cells; the hamathecium consists of apically free, persistent paraphyses and periphyses that line the ostiolar canal; the ascal apex has a pronounced, J- apical annulus; ascospores are ellipsoidal to fusiform, generally hyaline, 1-3 transversely septate and germinate by a germ tube. The anamorphs exclusively have the enteroblastic-phialidic conidiogenesis and belong to several genera of dematiaceous hyphomycetes.

Although the generic concept of *Chaetosphaeria* seems to be clear, at present the genus includes species differing in some important diagnostic characters that are incompatible with those given by Tulasne & Tulasne (1863) and by Booth (1957, 1958). Fuckel (1870) extended the original concept of *Chaetosphaeria* and included two species, *C. phaeostroma* (Durieu & Mont.) Fuckel and *C. fusca* Fuckel, that have superficial, obpyriform, setose perithecia seated on a brown hyphal mat, 3-septate ascospores, with the two central cells becoming dark brown and the two terminal cells remaining hyaline. Fuckel was mistaken in placing them in *Chaetosphaeria*. It was also a mistake to place them in *Thaxteria* Speg. (Booth, 1958). Saccardo (1883) included in *Chaetosphaeria* species possessing superficial or semi-immersed, black perithecia that are glabrous or covered by light coloured hairs and that have several-celled, brown ascospores. Fuckel´s (Fuckel, 1870) and Saccardo´s (1883) concept of *Chaetosphaeria* was followed by other authors (Winter, 1887; Schröter, 1889; Ellis & Everhart, 1892; Höhnel, 1909; Petch, 1917; Hughes, 1966) who extended the generic concept beyond the point of usefulness. Within the genus they included species with dark or light coloured, setose or nonsetose perithecia; perithecia with or without a subiculum or that are seated on a thin hypostroma; ascospores hyaline, dark brown or versicoloured; anamorphs producing holoblastic or enteroblastic-phialidic conidia. This generic concept proved to be very heterogenous.

The preliminary revision of some *Chaetosphaeria* species (Réblová, 1997) revealed that some species with dark-coloured ascospores had bitunicate asci often belong to genera of the Dothideales, e.g. *Byssosphaeria* Cooke and *Pseudotrichia* Kirschst. It was also proven that not all species included in *Chaetosphaeria* and described as possessing hyaline, several-septate ascospores and unitunicate asci, are correctly placed in the genus (Réblová, 1997). Some taxa having versicoloured ascospores have already been transferred from *Chaetosphaeria* to two new genera, viz. *Melanochaeta* E. Müll., Harr & Sulmont (Müller & al., 1968) and *Chaetosphaerella* E. Müll. & C. Booth (Müller & Booth, 1972). Nevertheless, *Chaetosphaeria* s. str. (Tulasne & Tulasne, 1863; Booth, 1957, 1958) remains as a large, aberrant taxon whose species are very similar in morphology of the perithecium, ascus, ascospore and hamathecium. The great diversity within the genus is demonstrated by the large number of associated anamorphs, belonging to eleven dematiaceous hyphomycete genera, producing enteroblastic-phialidic conidia.

Chaetosphaerella was separated from *Chaetosphaeria* (Müller & Booth, 1972) by perithecia that are superficial on a dense subiculum, obpyriform, tuberculate, collapsing inwards when dry, stipitate, glabrous and provided with long, spine-like setae at the base; a wide, leathery peridium of angular, thick-walled cells that form a pallisade at the surface of the perithecia, the wall cells at the exterior tending to be very thick-walled and opaque; a thin-walled, J- ascal apex that lacks any visible discharge mechanism; broadly cellular, deliquescent paraphyses; oblong, 3-septate, versicoloured ascospores; synanamorphs with holoblastic-tretic and eneteroblastic-phialidic conidiogenesis.

Lasiosphaeria (Cesati & de Notaris, 1863) is clearly distinct from both *Chaetosphaeria* and *Chaetosphaerella* in having perithecia that are superficial or partly immersed, pyriform to conical, glabrous or setose or with a thin hyphal cover, distinctly tuberculate or smooth; a peridium leathery to partly carbonaceous that comprises angular, thick-walled cells; an ascal apex with a subapical chamber and plasmatic globule, a pronounced, J-apical annulus; filiform paraphyses; ascospores are cylindrical, bent in the lower third, 0-multi-septate, hyaline, sometimes becoming yellowish to pale brown with age and germinate by both germ tube and phialide; known anamorphs that have enteroblastic-phialidic conidiogenesis and that belong to *Phialophora* Medlar. For the history of *Lasiosphaeria* see Lundqvist (1972).

Several criteria are suggested to have diagnostic value in distinguishing the twenty-eight species having versicoloured ascospores; viz. type of ascospore (colour, shape, septation), type of ascal apical opening (absence or presence, appearance, the amyloid or nonamyloid reaction), character of paraphyses (persistent or deliquescent, appearance) and conidiogenesis. The peridium-type is another important criterion and its anatomy is characteristic for several genera (e.g. *Chaetosphaeria*, *Chaetosphaerella*, *Helminthosphaeria*, *Melanochaeta*). Usually the peridium type is correlated with a particular ascospore-type, with the exception of *Lasiosphaeria*, a large, rather heterogenous genus that requires a critical revision and where several peridium-types and several ascospore-types are present. According to Carrol & Munk (1964) the genus shows a marked lack of correlation of characters.

Due to the correlation of diagnostic characters observed in type and other herbarium material of these species they are distributed among eight genera and three families. Three previously known genera are accepted, namely *Chaetosphaerella* and *Miyoshiella* Kawamura of the Helminthosphaeriaceae and *Melanochaeta* (Réblová, 1997) of the Chaetosphaeriaceae. One new genus is introduced in the Helminthosphaeriaceae and three new genera are introduced to the Trichosphaeriaceae. These are discussed in the respective parts of this series of publications.

The present paper deals with species of the genus *Chaetosphaerella*. Four species were described in the genus, viz. *Chaetosphaerella fusca* (Fuckel) E. Müll. & C. Booth, *C. fusispora* Sivan., *C. indica* Varghese & V.G. Rao and *C. phaeostroma* (Durieu & Mont.) E. Müll. & C. Booth. Type, exsiccata and other material of these species and that of additional four species, viz. *Chaetosphaeria bihyalina* (Berk. & Broome) Sacc., *C. incrustans* Rick, *C. iquitosensis* (P. Henn.) Theiss. and *C. phaeostromoides* (Peck) Sacc., was examined. Two species are accepted in the genus, namely *Chaetosphaerella fusca* and *C. phaeostroma*. *Chaetosphaeria incrustans*, *C. bihyalina* and *C. phaeostromoides* are synonyms of *C. fusca*. On the basis of the peridium anatomy, character of setae, ascal anatomy and conidiogenesis *Chaetosphaerella* is assigned to the Helminthosphaeriaceae. All accepted species of *Chaetosphaerella* are listed with full synonymy along with notes on previous descriptions and illustrations. The relationship of *Chaetosphaeria iquitosensis* with *Chaetosphaerella* is discussed.

Based on the investigations of type and other herbarium material of *Chaetosphaerella fusispora* and *Chaetosphaerella indica* neither proved to be correctly placed in the genus and both are excluded from *Chaetosphaerella*. The examination of *Chaetosphaerella indica* revealed that the fungus has morphological characters of perithecium and ascus and a conidiogenesis suggesting its affinity with the Helminthosphaeriaceae (Samuels & al., 1997) but it does not fit into any known genus of that family. Therefore, a new genus *Tengiomyces* is described with the single species *T. indicus*, for fungi characterized by superficial, globose to subglobose, nonpapillate, setose perithecia that are collabent in the upper part when dry, a truncate perithecial apex that is formed of more or less clavate, thick-walled cells; early deliquescent, cellular paraphyses; thick-walled asci, ascal apex without any visible discharge mechanism; 3-septate, ellipsoidal, versicoloured ascospores. Conidiogenesis of the presumed *Spadicoides* S. Hughes anamorph is holoblastic-tretic.

The revision of *Chaetosphaerella fusispora* showed that the fungus is distinct from *Chaetosphaerella* and the Helminthosphaeriaceae in the characters of peridium anatomy and hamathecium and ascal anatomy. Its affinity lies with the Trichosphaeriaceae where it is placed in a new genus (Réblová, 1998b).

MATERIALS AND METHODS

Dry herbarium specimens were rehydrated in 3% KOH and subsequently studied in 100% lactic acid, water, Congo Red (aq.) or Melzer´s reagent. The structure of the perithecial wall was studied from freehand sections of dried material cut under the dissecting microscope and mounted in lactic acid. Two types of microscopy were used in this study. These are indicated in the legends to the illustrations as bright field (BF) and phase contrast (PC). Photographs were taken in water, Congo Red (aq.) or in Melzer´s reagent. Abbreviations of the herbaria and institutes which kindly lent material are cited in accordance with Index Herbariorum (Holmgren & al., 1990).

TAXONOMIC PART

Chaetosphaerella E. Müll. & C. Booth, Trans. Brit. Mycol. Soc. 58: 76, 1972.

Perithecia uniform in shape, obpyriform with a small papilla at the top, collapsing inwards when dry, attached to the substratum by a sterile, stout base, seated in a dark subiculum of the associated anamorph, distinctly tuberculose in *C. fusca* and only slightly roughened in *C. phaeostroma*, glabrous, at the base provided with a few erect, dark, opaque, thick-walled, acute, sterile setae which are also scattered among conidiophores of *Oedemium minus* (Link) S. Hughes the anamorph of *C. phaeostroma.*—Peridium consisting of two layers, the outer layer of thick-walled, polyhedral cells, the inner layer of thin-walled, nonpigmented, flattened cells, the sterile perithecial base formed of divergent rows of vertically arranged cells. A palisade of cylindrical cells, heavily pigmented on the exterior, is arranged on the outer side of the peridium.—Ostiolar canal periphysate.—Paraphyses of broadly cellular, inflated segments, present only in early stages of the ascoma development, deliquescing early and forming an amorphous matrix among the mature asci. The amorphous matrix, as well as the young asci, stained slightly by Congo Red; mature asci not staining in Congo Red.—Asci arising in a broad hymenium, cylindric, shortly stipitate, rounded at the top; apex with an indistict, J- apical annulus.—Ascospores 4-celled, oblong, the two middle cells longer, becoming brown, the two end cells remaining hyaline, slightly constricted or not constricted in the middle. In *C. fusca* one of the middle cells may become slightly inflated.

The anamorph belongs to the hyphomycete genus *Oedemium* (Hughes & Hennebert, 1963). *Chaetosphaerella phaeostroma* has *Oedemium minus* and *Veramycina elegans* Subram. synanamorphs (Subramanian, 1993), the two anamorphs differ in conidiogenesis,viz. holoblastic, forming solitary or catenate conidia from percurrently proliferating conidiogenous cells in the former and enteroblastic in the latter.

TYPE SPECIES. *Chaetosphaerella phaeostroma* (Durieu & Mont.) E. Müll. & C. Booth (Müller & Booth, 1972).

SYNANAMORPHS. *Oedemium* Link, in Linné´s Sp. Plant., Ed. 4 (Willdenow´s), 6: 42-43, 1824.

Veramycina Subram., Kavaka 20-21: 58, 1995.

≡ *Veramyces* Subram., Czech Mycol. 47: 4, 1993. (Not validly published. Art. 37.1.) non *Veramyces* Matsush. 1993.

SYSTEMATIC POSITION. Helminthosphaeriaceae Samuels, Candoussau & Magni, Trichosphaeriales M.E. Barr.

ACCEPTED TAXA. *Chaetosphaerella fusca* (Fuckel) E. Müll. & C. Booth, *C. phaeostroma* (Durieu & Mont.) E. Müll. & C. Booth.

HABITAT. Saprobes on woody substrata and old stromata of some species of the Diatrypaceae.

KNOWN DISTRIBUTION. Probably widespread in temperate and tropical regions and known from Asia (Sri Lanka), Europe (Czech Republic, France, Germany, Great Britain, Italy, Russia), North America (U.S.A.) and South America (Brazil).

Key to species of *Chaetosphaerella*

1. Ascospores usually more than 30 μm long, perithecia associated with *Oedemium minus* anamorph .... *C. phaeostroma*

1. Ascospores usually less than 30 μm long, perithecia associated with *Oedemium didymum* anamorph .... *C. fusca*

1. ***Chaetosphaerella fusca*** (Fuckel) E. Müll. & C. Booth, Trans. Brit. Mycol. Soc. 58: 77, 1972. Figs. 1-11, 16.

≡ *Chaetosphaeria fusca* Fuckel, Jahrb. Nassau. Ver. Naturk. 23-24: 166, 1870.

≡ *Thaxteria fusca* (Fuckel) C. Booth, Naturalist 1958: 90, 1958.

= *Sphaeria bihyalina* Berk. & Broome, J. Linn. Soc. Bot. 14: 124, 1873.

≡ *Chaetosphaeria bihyalina* (Berk. & Broome) Sacc., Syll. Fung. 2: 93, 1883.

= *Sphaeria phaeostromoides* Peck, New York State Mus. Rep. 28: 77, 1876.

≡ *Chaetosphaeria phaeostromoides* (Peck) Sacc., Syll. Fung. 2: 93, 1883.

≡ *Chaetosphaeria phaeostroma* (Durieu & Mont.) Fuckel var. *phaeostromoides* (Peck) Ellis & Everh., North Amer. Pyrenomyc., p.160, 1892.

= *Chaetosphaeria incrustans* Rick, Broteria 3: 289, 1904.

ANAMORPH. **Oedemium didymum** (Link) S. Hughes, Canad. J. Bot. 36: 790, 1958. Fig. 11.

Perithecia superficial, densely gregarious, obpyriform, ostiolate, 230-270(-350) µm wide and 390-510 µm high, collapsing inwards when dry, attached to the substratum by a sterile obconic base, dark brown to black, glistening, deeply tuberculose, sitting on a dense, reddish-brown hyphal mat of sterile hyphae and conidiophores of the anamorph. Perithecia at the base provided with sterile, dark brown, septate, tapering, straight or slightly flexuous, unbranched or seldom dichotomously branched setae up to 180 µm long and 3-4 µm wide in the middle, with blunt to slightly pointed apicies.—Peridium leathery, 40-57 µm wide, consisting of two layers. Outer layer 33-44 µm wide, of thick-walled, brown, polyhedral cells with Munk pores in the wall. Cells on the outer side of the wall cylindrical, heavily pigmented at the exterior and arranged in a palisade forming tubercules on the surface. Inner layer 7-17 µm wide, of thin-walled, nonpigmented, elongated, compressed cells. Inner cells of the stalk thick-walled, polyhedral to cylindrical, arranged in divergent vertical rows.—Ostiolar canal periphysate.—Paraphyses of broadly cellular, inflated segments 15-24 µm wide and 10-55 µm long in their lower part, and smaller towards the apex, 5-11 µm wide and 11-17 µm long, deliquescing early, present in young perithecia only.—Asci unitunicate, 8-spored, 80-100 × 15-16(-17) µm, cylindrical, short stipitate, apex J-, with an indistinct apical annulus.—Ascospores oblong, straight to slightly curved, (21-)23-31(-34) × (4.5-)5-7(-8.5) µm, 4-celled, the two longer middle cells brown, the two terminal cells remaining hyaline to subhyaline, not constricted or slightly constricted at the middle septum, 1-2-3-seriate, partially overlapping in the ascus. One of the middle cells may become slightly inflated.

MATERIAL EXAMINED. 1) Exsiccata. Petrak, Flora Bohem. & Morav. Exsic. No. 676, Lfg. 14 (CZECH REPUBLIC, Moravia, Hrabůvka near Hranice na Moravě, on branch of a deciduous tree, March 1913, F. Petrak, PRM 656909). - Saccardo, Mycotheca italica No. 303 (ITALY, Padova, botanical garden, on the base of a trunk of *Morus papyrifera*, Jan. 1898, P. A. Saccardo, PRM 656905). - Saccardo, Mycotheca Veneta No. 679 (ITALY,

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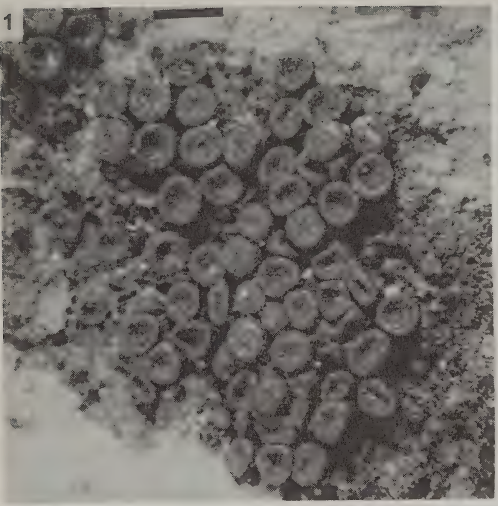
Figs. 1-6. **Chaetosphaerella fusca**. - 1. Perithecia sitting on subiculum. - 2. Longitudinal section of the perithecium center. - 3. Median, longitudinal section of the upper part of perithecium. - 4. Vertical rows of cells of stalk. - 5. Asci. - 6. Ascospore. - Figs. 1-6: BF. - Fig 1 from K 37398; Figs. 2, 3, 4, 6 from PACA 12664; Fig. 5 from PACA 12669. - Scale bars: 1 = 500 µm; 2, 3 = 100 µm; 4, 5 = 20 µm; 6 = 10 µm.

1



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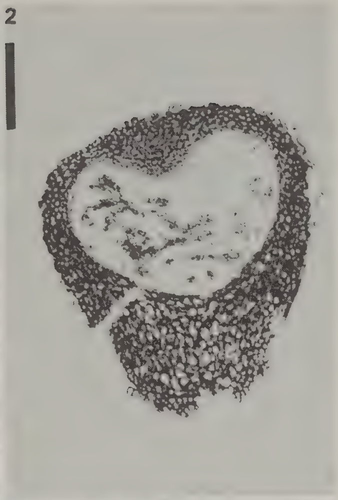

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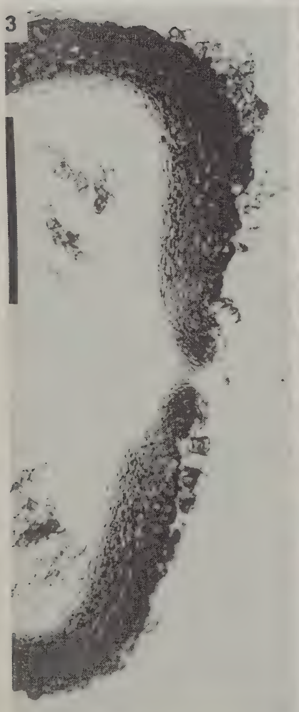

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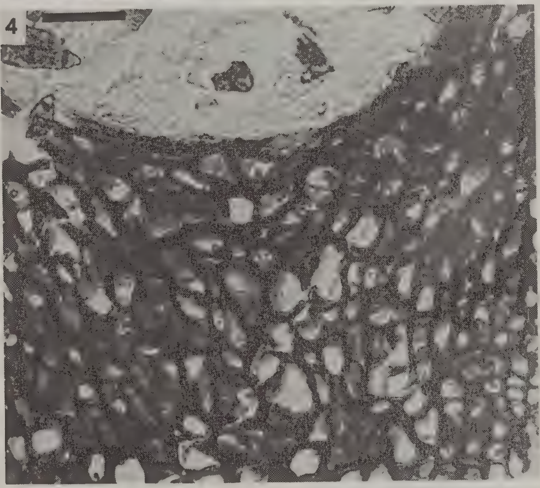

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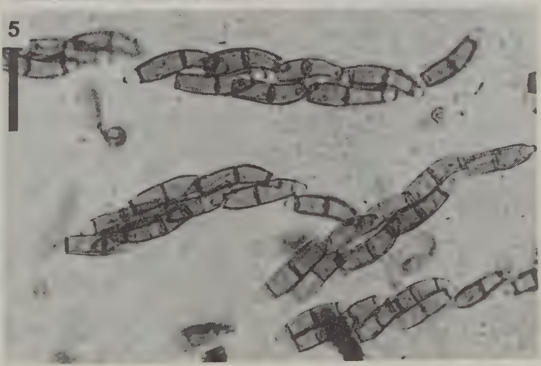

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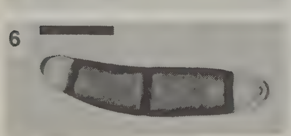

Padova on decayed trunk of *Robinia pseudoaccacia* and *Broussonetia*, March 1873, P.A. Saccardo, W); No. 660 (ITALY, Selva (Treviso) on trunk of *Ulmus* and *Broussonetia*, Sept. 1875, P.A. Saccardo, W); No. 1453 (Italy, Padova, on a branch, Bizzozer, Nov. 1880, W). - Rick, Fungi austro-americani No. 107 (BRAZIL, São Leopoldo, on wood of a deciduous tree, 1905, J. Rick, PACA 12679, PACA 12673).

2) Type material. Fuckel, Fungi rhen. No. 2041 (G - holotype of *Chaetosphaeria fusca*) - SRI LANKA. Central Province, on dead undetermined wood, July 1868, Thwaites, No. 560 (K 37398 - holotype of *Sphaeria bihyalina*).

3) Additional material. CZECH REPUBLIC. Central Bohemia, Škvorecká obora, on bare wood of a trunk of *Sorbus aucuparia*, 28 March 1982, M. Svrček (PRM). - BRAZIL. São Leopoldo, on wood of deciduous tree, 1906, J. Rick (PACA 12664); ibid., 1929, J. Rick (PACA 12669). - FRANCE. Massif Central Mts., Tanus near Rodez, on decorticated branch of *Quercus robur*, 12 July 1997, M. Réblová (Herb. M. Réblová 922/97). - U.S.A. Six Mile Ravine near Ithaca, Tompkins County, on a rotten log, 25 Sept. 1948, C. T. Rogerson (BPI 622130).

DESCRIPTIONS AND ILLUSTRATIONS. Fuckel (1870: 166); Berkeley & Broome (1873: 124); Saccardo (1883: 93); Schröter (1889: 309); Peck (1876: 77); Winter (1887: 219); Berlese (1890: Tab. 17, Fig. 6); Ellis & Everhart (1892: 160); Petch (1917: 331); Petrak (1921: 276); Booth (1958: 90, Fig. 4m); Ellis (1971a: 176, Fig. 119a).

HABITAT. Saprobe on bark and decorticated wood.

KNOWN HOSTS. *Acer pseudoplatanus, Alnus glutinosa, Broussonetia* sp., *Corylus avellana, Fagus sylvatica, Morus papyrifera, Quercus robur, Robinia pseudoaccacia, Sorbus aucuparia, Salix* sp., *Tilia* sp. and *Ulmus* sp.

KNOWN DISTRIBUTION. The fungus is widespread in tropical and temperate regions. I have examined material from the Czech Republic, Brazil, France, Germany, Italy, Sri Lanka and the U.S.A.

Figs. 7-15. 7-11. ***Chaetosphaerella fusca***. 12-15. ***Chaetosphaerella phaeostroma***. - 7. Longitudinal section of perithecium. - 8. Ascus. - 9. Ascospores. - 10. Asci. - 11. Conidiophores and conidia of the ***Oedemium didymum*** anamorph, from nature. - 12. Longitudinal section of perithecium. - 13. Conidiophores of the ***Veramycina elegans*** synanamorph, from nature. - 14. Median, longitudinal section of the upper part of perihecium. - 15. Ascoma. - Figs. 7-15: BF. - Figs. 7-11 from Herb. M. Réblová 922/97; 12-15 from Herb. M. Réblová 1139/97. - Scale bars: 7, 8, 12, 15 = 100 µm; 9 = 10 µm; 10, 14 = 50 µm.

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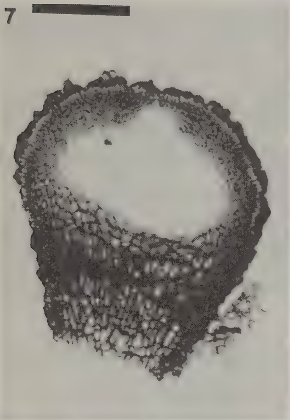

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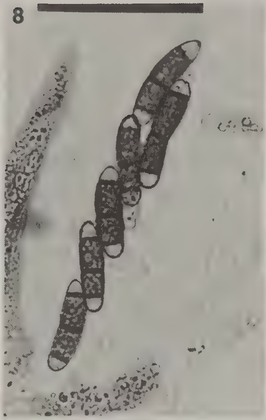

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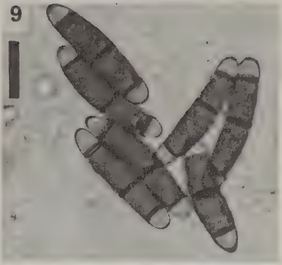

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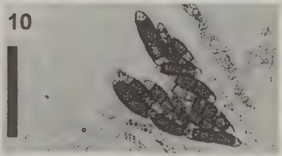

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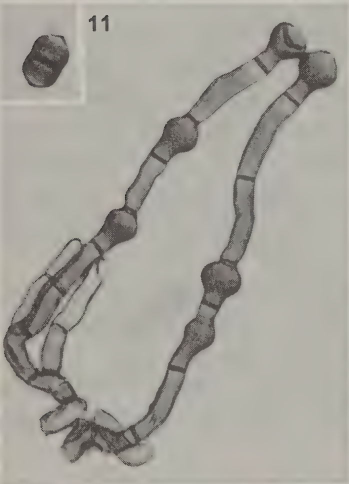

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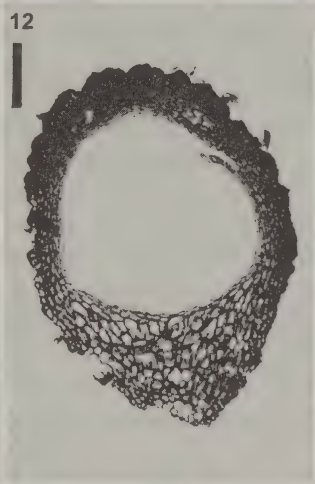

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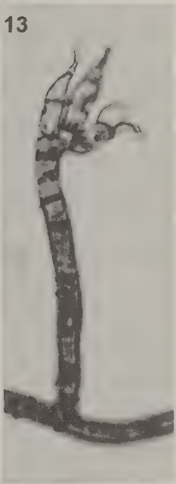

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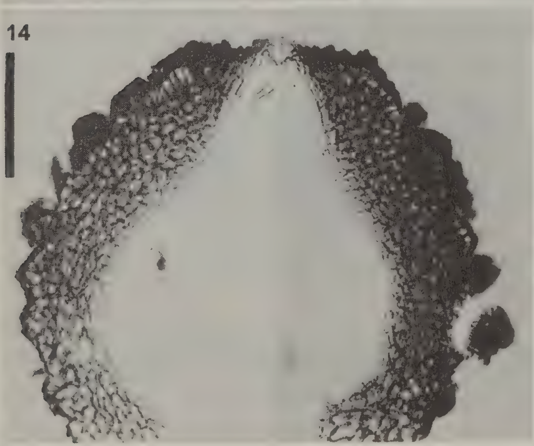

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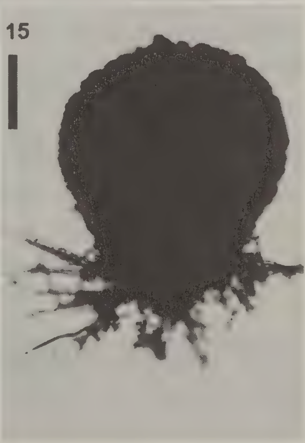

Other reliable reports come from Germany (Fuckel, 1870; Winter, 1887), Great Britain(Booth, 1958) and France (Saccardo, 1883). It is also mentioned from Russia (Potebnia, 1910).

Sterile setae growing from the base of the perithecia differ in habit from those of *Chaetosphaerella phaeostroma*; they are finer, more flexuous, almost blunt at the top and are not scattered among conidiophores of the anamorph as is typical of *C. phaeostroma* where the setae are spine-like.

The palisade of cylindrical cells with heavily pigmented outer wall arranged at the surface of perithecia is most conspicuous in young perithecia. The palisade of cells was observed to be less prominent or even lacking in collections made by Rick in Brazil.

Petch (1917) revised the type collection of *Sphaeria bihyalina*. His observations agree well with mine.

Berlese (1890) regarded *Chaetosphaerella fusca* as a synonym of *Chaetosphaeria bihyalina*. His description and illustration are based on the specimen from Saccardo´s herbarium. The fungus is described as having hairy perithecia 250 µm wide, asci 60-70 x 10-12 µm and ascospores 18-22 x 6-7 µm. It is obvious that Berlese´s fungus and that collected in Sri Lanka are not identical. The hyphomycetous species, which is described and illustrated by Berlese as accompanying the perithecia, probably belongs to the genus *Helminthosporium* and is not connected to the perithecia. Saccardo´s specimen has not been examined by me.

The type material of *Chaetosphaeria incrustans* Rick is probably lost. The only surviving original element is its description. The fungus already has been designated by Rick as a synonym of *Chaetosphaeria bihyalina* (Rick, Broteria 5: 136, 1906). Judging from the protologue it seems that Rick´s opinion is correct, and the name is listed as a synonym of *Chaetosphaerella fusca*.

Chaetosphaerella fusca occurs seldom, but is more frequent as an anamorph. The anamorph is a lignicolous saprobe found growing together with the perithecia or independently on the wood and bark of different kinds of trees. Illustrations and descriptions are provided by Ellis (1971a: 176, Fig. 119a), Hughes & Hennebert (1963: 803, Figs.1, 2, 18-20) and Booth (1958: 90, Figs. 4n). Ellis (1971a) recorded *Oedemium didymum* on fallen branches of various deciduous trees, e.g. *Acer, Fagus, Ilex, Salix* and *Tilia*.

2. ***Chaetosphaerella phaeostroma*** (Durieu & Mont.) E. Müll. & C. Booth, Trans. Brit. Mycol. Soc. 58: 77, 1972.

Figs. 12-15, 17, 18.

≡ *Sphaeria phaeostroma* Durieu & Mont., Expl. Sci. Algérie, p. 491, 1849.

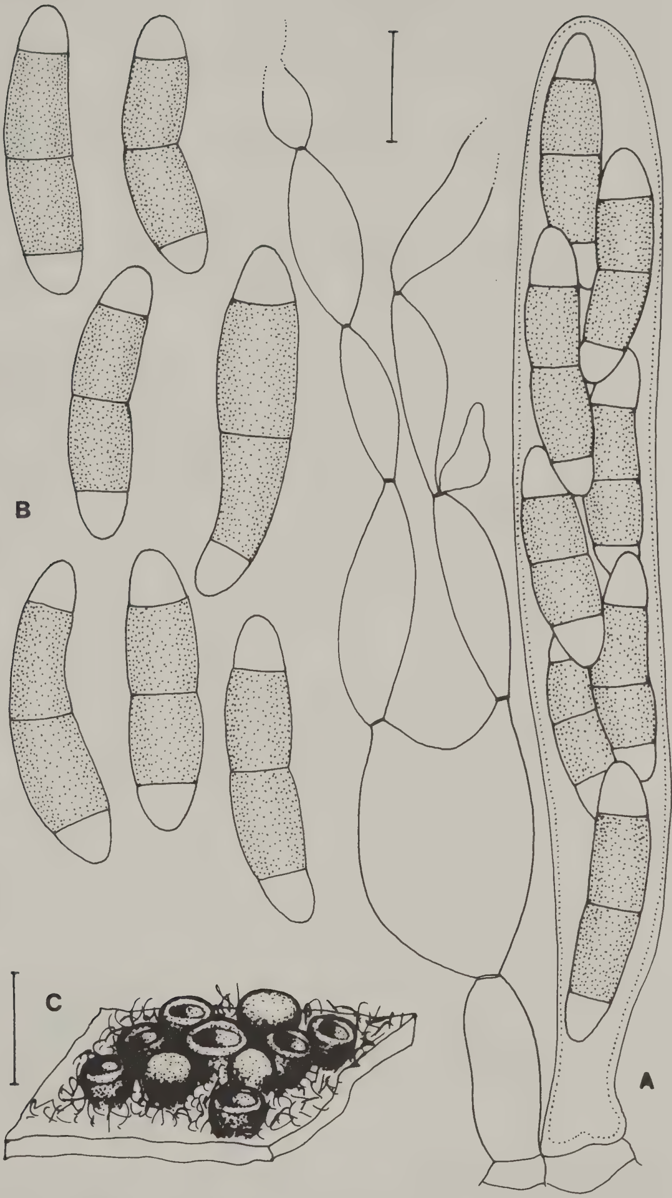

Fig.16A-C. ***Chaetosphaerella fusca***. - A. Asci, ascospores and paraphyses. - B. Ascospores. - C. Habit sketch of perithecia sitting in a subiculum. - A-C from Herb. M. Réblová 922/97). - Scale bars: A, B = 10 µm; C = 500 µm.

≡ *Chaetosphaeria phaeostroma* (Durieu & Mont.) Fuckel, Jahrb. Nassau. Ver. Naturk. 23-24: 166, 1870.

≡ *Thaxteria phaeostroma* (Durieu & Mont.) C. Booth, Naturalist 1958: 88, 1958.

= *Chaetosphaeria tristis* (Tode: Fr.) Schröter sensu Schröter, Die Pilze Schlesiens 3/2: 308, 1908, non *Sphaeria tristis* Tode: Fr., Syst. Mycol. 2: 444, 1823.

SYNANAMORPHS. ***Oedemium minus*** (Link) S. Hughes, Canad. J. Bot. 36: 790, 1958. ***Veramycina elegans*** Subram., Kavaka 20-21: 58, 1995. Figs. 13, 18.

Perithecia superficial, densely gregarious, obpyriform, ostiolate, 280-350 µm wide and 350-480 µm high, seldom collapsing inwardly when dry, attached to the substratum by a sterile obconic base, dark brown to black, glistening, slightly tuberculose, seated on a blackish subiculum of sterile hyphae, conidiophores and sterile anamorphic setae. Similar setae growing from the base of the perithecia; setae spine-like, dark brown, opaque, stiff, erect, acute, unbranched or dichotomously branched, up to 350 µm long and 7-8.5 µm wide in the middle.—Peridium leathery, 39-55 µm wide, consisting of two layers. Outer layer 32-49 µm wide, of thick-walled, brown, polyhedral cells with Munk pores in the wall. Cells on the outer side of the wall cylindrical, heavily pigmented on the exterior and arranged in a palisade forming low tubercules at the surface. Inner layer 4.5-6.5 µm wide of thin-walled, nonpigmented, elongated, compressed cells. Inner cells of the stalk polyhedral to cylindrical, arranged in divergent vertical rows.—Ostiolar canal periphysate.—Paraphyses of broadly cellular, inflated segments 17-29 µm wide and 26-66 µm long in the lower part, becoming smaller towards the apex, 4-6 µm wide and 13-18 µm long in the upper part, deliquescing early, present in young perithecia only.—Asci unitunicate, 8-spored, 100-124 × 16-23 µm, cylindrical-clavate, short-stipitate, apex with an indistinct, J- apical annulus.—Ascospores oblong, straight or slightly curved, (29-)30-40(-44) × 7-10.5(-11.5) µm, 4-celled, the two longer middle cells brown and the two end cells remaining hyaline, not constricted or slightly constricted at the middle septum, obliquely uniseriate or 2-3-seriate, partially overlapping in the ascus.

MATERIAL EXAMINED. 1) Exsiccata. Petrak, Mycotheca carpatica Exsic. No. 178 (CZECH REPUBLIC, Moravia, Carpathian Mts., Javornik Mt., on old wood, 19 July 1918, F. Petrak, PRM 656908). - Petrak, Mycotheca carpatica Exsic. No. 78 (CZECH REPUBLIC, Moravia, Beskydy Mts.,

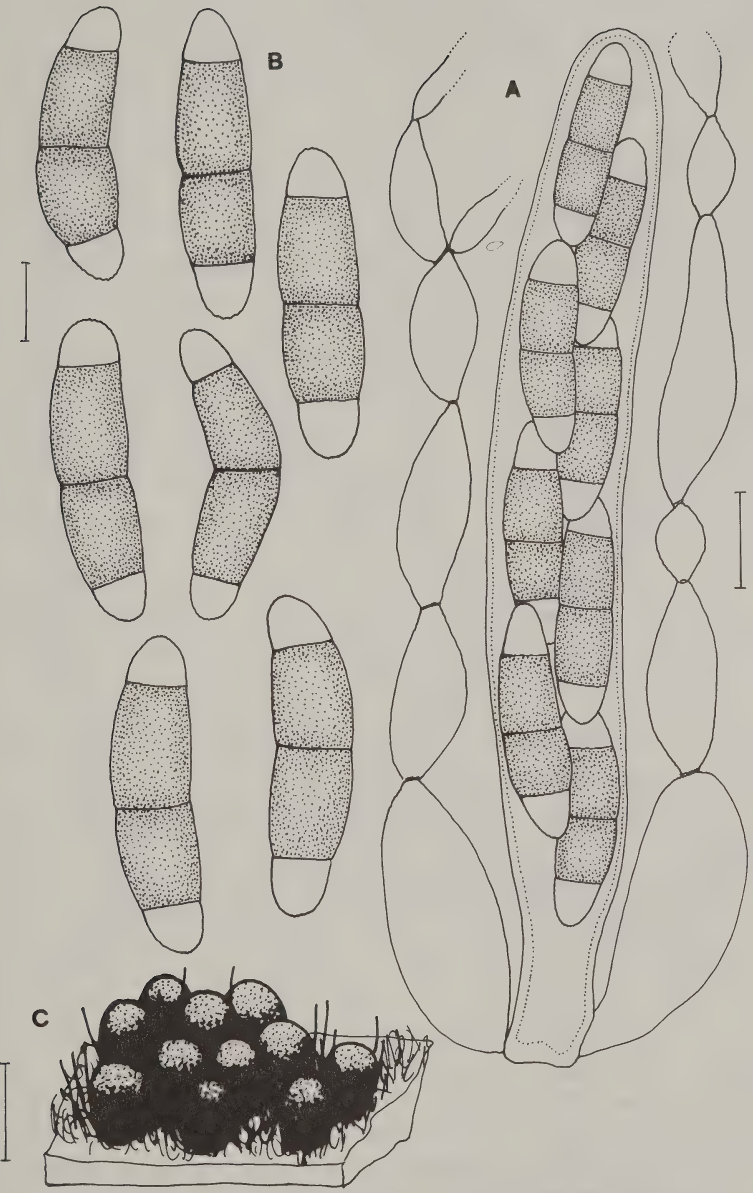

Fig. 17A-C. ***Chaetosphaerella phaeostroma***. - A. Asci, ascospores and paraphyses. - B. Ascospores. - C. Habit sketch of perithecia sitting in a subiculum. - A-C from PRM 659908. - Scale bars: A, B = 10 µm; C = 500 µm.

Rožnov, on old wood of a deciduous tree, 28 Sept. 1920, F. Petrak, PRM 656907). - Saccardo, Mycotheca italica Exsic. No. 645 (ITALY, on bark of *Picea abies*, June 1899, PRM 656906). - Fuckel, Fungi rhen. No. 2040 (G).

2) Additional material. CHINA. Yunnan, Kai-yuan, on bare wood, 21 March 1934, Y. Tsiang (BPI 622129). - CZECH REPUBLIC. Central Bohemia, on the slopes of Mt. Hradiště (369 m a.s.l.) near Praha, on old stroma of *Eutypa flavovirens* on *Carpinus betulus*, 8 Sept. 1996, M. Réblová (Herb. M. Réblová 866/96). - ibid., Malá Chuchle near Praha, on stump and branch of *Quercus petraea*, 12 April 1964, V. Holubová-Jechová (Herb. M. Réblová 884/97). - ibid., Budňany, in the valley of the river Berounka, on strongly decayed wood of a deciduous tree, 1 March 1953, M. Svrček (PRM). - ibid., Dobřichovice, on the slopes of Mt. Červená hlína (467 m a.s.l.), on a branch of *Carpinus betulus*, 19 July 1990, M. Svrček (PRM). - ibid., on a trunk of *Alnus glutinosa,* 19 July 1990, M. Svrček (PRM). - ibid., Chvojen, on a branch of *Corylus avellana*, 10 June 1978, M. Svrček (PRM). - Southern Bohemia, Šumava Mts., Modrava, Medvědí hřbet below Mt. Roklan, on bark of *Acer pseudoplatanus*, 10 June 1997, Z. Palice (Herb. M. Réblová 1139/97).

DESCRIPTIONS AND ILLUSTRATIONS. Fuckel (1870: 166); Saccardo (1883: 93); Winter (1887: 218; 195: Figs. 1-4); Berlese (1890: 27, Pl. 17, Fig. 5); Schröter (1889: 308); Teng (1934: 363, Fig. 6); Booth (1958: 88, Fig. 4o); Müller & Booth (1972: 77, Figs. 1, 3b-e).

HABITAT. Saprobe on bark and decorticated wood and also often growing on old stromata of species in the Diatrypaceae, most commonly on those of *Diatrype stigma* and *Eutypa flavovirens*.

KNOWN HOSTS. *Acer pseudoplatanus, Broussonetia* sp., *Carpinus betulus, Corylus avellana, Fagus sylvatica, Fraxinus excelsior, Hedera helix, Ilex, Lonicera xylosteum, Picea abies, Pistacia lentiscus, Prunus* (*P. spinosa*), *Quercus* (*Q. petraea*), *Ribes, Salix* sp., *Tilia* sp., *Ulmus* (*U. americana*).

KNOWN DISTRIBUTION. Commonly collected in temperate regions of Europe and North America but also reported from China (Teng, 1934) and French Guiana (Courtecuisse & al., 1996). I have examined material from China, the Czech Republic and Italy. Other records include ones from Austria (Strasser, 1911), Denmark (Munk, 1957), France (Saccardo, 1883), Germany (Fuckel, 1870; Winter, 1887), Great Britain (Booth, 1958) and Sweden (Eriksson, 1992). Other collections were made in North America, South Carolina and Missouri, (Ellis & Everhart, 1892; Farr & al., 1989).

NOMENCLATURAL NOTE. *Sphaeria tristis* Tode was introduced by Tode (Fungi Mecklenb. Sel. 2: 9, Tab. IX, Fig. 67, 1791) for an ascomycetous fungus described as "simplex, aggregata, atra, immersa, lichenoides, minima granulata" and illustrated with subglobose, half

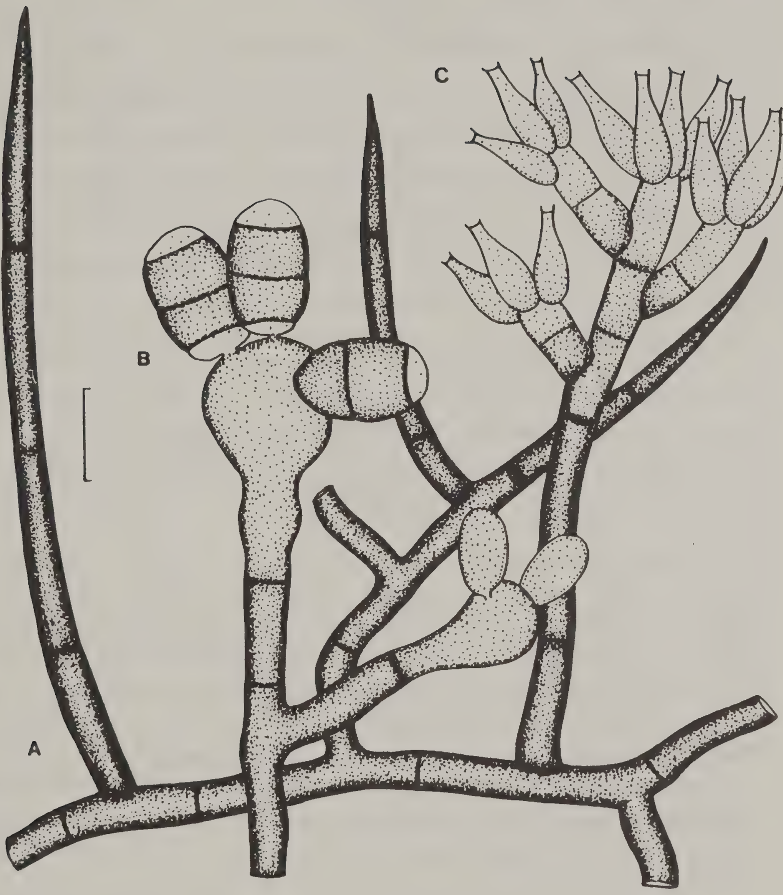

Fig. 18A-C. ***Chaetosphaerella phaeostroma***. - A. Seta of the subiculum. - B. Conidiophore with conidia of the ***Oedemium minus*** synanamorph, from nature. - C. Conidiophore of ***Veramycina elegans*** synanamorph, from nature. - A-C from Herb. M. Réblová 866/96). - Scale bars: A-C = 20 μm.

immersed perithecia surrounded by a dense hyphal mat. No type material of *Sphaeria tristis* is available as none of Tode´s original collections are preserved. The illustration accompanying the original description is the only surviving original element. Unfortunately, this illustration could not be identified with any subsequent collection.

Persoon (Icon. et descript. Fung. cognitorum, p. 49, Tab. XII, Figs. 5, 6, 1800; Synop. Meth. Fung., p. 87, 1801) used Tode´s name *Sphaeria tristis* in his own new interpretation. In his earlier work of 1800, the author did not fully agree with the Tode´s description and added to it his personal observations and an illustration which is quite different from that of Tode. Persoon described the fungus as "simplex conferta atra crustaeformis, sphaerulis subglobosis, utplurimum collapsis, tomento strigoso insidentibus" and he noted "omnino hujus loci videtur, verum, saltem meis in speciminibus, sphaerulae ipsae ligno non sunt immersae, nec orificio plano pertusae, quod hac de specie in descriptione 6. Todei adnotatum invenies".

Fries (Sv. Vet. Acad. Handl. 36, 2: 252, 1817) quoted both Tode´s and Persoon´s descriptions and noted that two different species were probably involved under this name. Fries (Syst. Mycol. 1: 444, 1823) listed Tode´s description first and Persoon´s second, but he signed Persoon´s with a mark of exclamation. Fries obviously based his description of *Sphaeria tristis* on that of Persoon.

Fuckel (Jahrb. Nassauischen Ver. Naturk. 23-24: 165, 1870) used the name *Sphaeria tristis* Tode sensu Pers. as a basionym for a new combination in the genus *Nitschkia* [*Nitschkia tristis* (Pers.: Fr.) Fuckel = *Nitschkia grevillei* (Rehm) Nannf.] and simultaneously excluded *Sphaeria tristis* Tode as representing a species related to *Chaetosphaeria* Tul. & C. Tul. sensu Fuckel (= *Chaetosphaerella* E. Müll. & C. Booth). Schröter (Die Pilze Schlesiens 3/2: 308, 1908) made a combination in *Chaetosphaeria* based on *Sphaeria tristis* Tode as *Chaetosphaeria tristis* (Tode) Schröter and the earlier name *Chaetosphaeria phaeostroma* (Durieu & Mont.) Fuckel [= *Chaetosphaerella phaeostroma* (Durieu & Mont.) E. Müll. & C. Booth] included in its synonymy.

Nannfeldt (1975) based the name *Acanthonitschkia tristis* (Pers.: Fr.) Nannf. on Persoon´s name *Sphaeria tristis* Pers.: Fr. and ascribed the epithet ‚*tristis*‘ to Persoon. According to Nannfeldt (1975: 57) ‚*Sphaeria tristis* Tode has no connection with any of the species that have later passed under this name and has been supposed to be *Chaetosphaerella phaeostroma*‘.

I cannot agree with Nannfeldt´s opinion that *Sphaeria tristis* Tode is related to *Chaetosphaerella phaeostroma*. Judging from the protologue and original illustrations of *Sphaeria tristis* Tode, it evidently has no connection to *Chaetosphaerella phaeostroma* (Durieu & Mont.) E. Müll. & C. Booth. It cannot be identified with any of the known species of *Chaetosphaerella*, so the identity of this fungus remains obscure. Persoon was the first to give an understanding of Tode´s name *Sphaeria tristis*. His material is preserved in L (L 910. 269-833!); it is determinable and it was designated as a lectotype by Nannfeldt (1975).

The names *Chaetosphaerella phaeostroma* and *Acanthonitschkia tristis* are extensively used for fungi frequently reported from the temperate zone of Europe. Both names include in their synonymy the name *Sphaeria tristis*. Because it will never be possible to know what Tode actually studied, I accept Nannfeldt´s treatment of *Sphaeria tristis* Pers.: Fr. To avoid confusion I herewith lectotypify the name *Sphaeria tristis* Tode with Tode´s original illustration and epitypify it with Persoon´s material (L 910. 269-833!). Persoon did not describe a new species, he thought he had Tode´s species. So, the species should be cited as *Sphaeria tristis* Tode: Fr.

Chaetosphaerella phaeostroma is easily distinguishable from the closely related *C. fusca* by the larger ascospores and asci and the *Oedemium minus* and *Veramycina elegans* synanamorphs.

Veramycina elegans was described on the basis of culture studies of *Oedemium minus* (Subramanian, 1993). Hughes & Hennebert (1963) observed the same fungus in a single collection (DAOM 87857, not seen) and suggested it to be *Phialocephala*-like. They described the fungus attached to sterile setae and to conidiophore initials and small groups of phialides were also found on perithecia initials. Conidiophores of the *Veramycina elegans* synanamorph were found among conidiophores of *Oedemium minus* from the same mycelium in the recent collection made on *Carpinus betulus* in the Czech Republic (Herb. M. Réblová 866/96).

The anamorph is a lignicolous saprobe commonly found growing together with the perithecia or forming independent glistening, blackish, velvety colonies on bark and decorticated wood. Often growing as a fungicolous hypersaprobe on old stromata of Diatrypaceous fungi. Illustrations and descriptions were provided by Booth (1958: 88, Fig. 4p) and Hughes & Hennebert (1963: 779, Figs. 17, 26, 27).

Tengiomyces Réblová, gen. nov.

Perithecia superficialia, solitaria vel gregaria, globosa usque subglobosa, non papillata, ostiolata, exsiccata parte superiore collabescentia, dense setosa, setis crasse tunicatis, hyphis subiculi sparsis insidentia; parietibus bistratosis, canale ostiolari periphysato. Hamathecium cum paraphysibus deliquescentibus. Asci unitunicati, 8-spori, cylindraceo-clavati, non amyloidei, annulo apicali indistincto. Ascosporae ellipsoideae, 3-septatae,

versicolores, cellulis centralibus brunnescentibus, cellulis apicalibus permanenter hyalinis.
Species typica. Tengiomyces indicus (Varghese & V. G. Rao) Réblová.

Perithecia superficial, solitary to gregarious, globose to subglobose, nonpapillate, ostiolate, collapsing by lateral pinching when dry in the upper part, densely setose, seated on a thin subiculum.—Peridium brittle, consisting of two layers.—Ostiolar canal periphysate.—Paraphyses early deliquescing.—Asci unitunicate, 8-spored, cylindrical-clavate, thick-walled, apex with an indistinct, J- apical annulus.—Ascospores ellipsoidal, 3-septate, versicoloured, the two middle cells becoming pale brown to greyish-brown and the two end cells remaining hyaline.

TYPE SPECIES. *Tengiomyces indicus* (Varghese & V. G. Rao) Réblová.

ANAMORPH. *Spadicoides* sp.

SYSTEMATIC POSITION. Helminthosphaeriaceae Samuels, Candoussau & Magni, Trichosphaeriales M.E. Barr.

ACCEPTED TAXON. *Tengiomyces indicus* (Varghese & V. G. Rao) Réblová.

ETYMOLOGY. In reference to the name of S. C. Teng, a mycologist who greatly contributed to the knowledge of the fungi of China and who collected one of the specimens treated in the present paper.

Tengiomyces indicus (Varghese & V. G. Rao) Réblová, comb. nov.
Figs. 19-32.

≡ *Chaetosphaerella indica* Varghese & V. G. Rao, Biovigyanam 5: 2, 1979. - basionym.

= *Chaetosphaeria innumera* Berk. & Broome ex Tul. & C. Tul. sensu S. Teng, Sinensia 4: 364, 1934 non *Chaetosphaeria innumera* Berk. & Broome ex Tul. & C. Tul., Sel. Fung. Carp. 2: 252, 1863.

ANAMORPH. ***Spadicoides*** sp. Figs. 28-31.

Perithecia superficial, solitary to gregarious, globose to subglobose, nonpapillate, ostiolate, collapsing by lateral pinching when dry in the upper part, dark brown, setose, 210-220 µm wide and 250-310 µm high, in the

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Figs. 19-22. ***Tengiomyces indicus***. - 19. Perithecial setae. - 20, 22. Perithecium. - 21. Median, longitudinal section of perithecium. - Figs. 19-22: BF. - Figs. 19-22 from BPI 622100. - Scale bars: 19 = 20 µm; 20, 22 = 100 µm; 21 = 50 µm.

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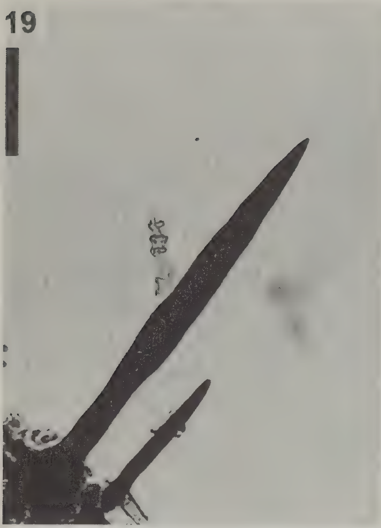

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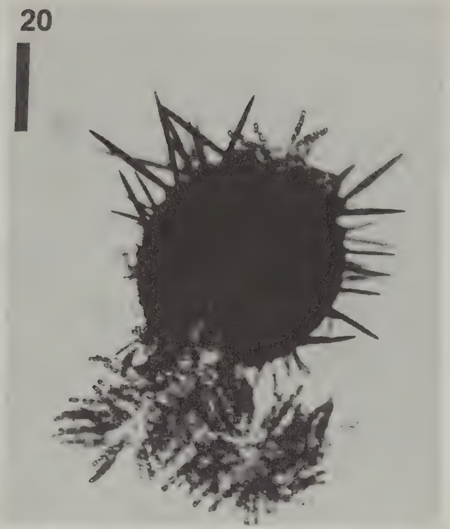

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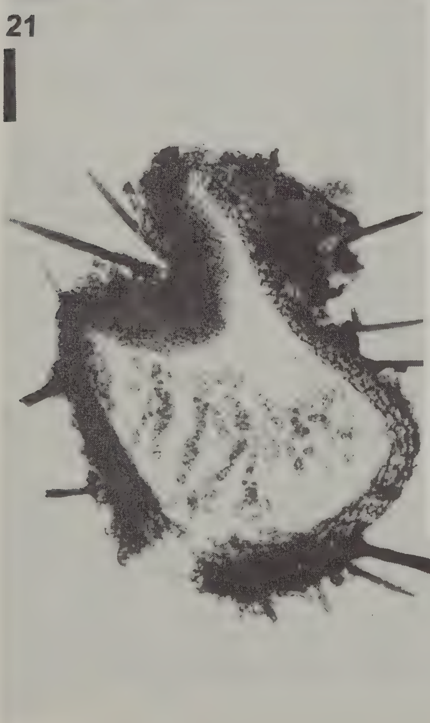

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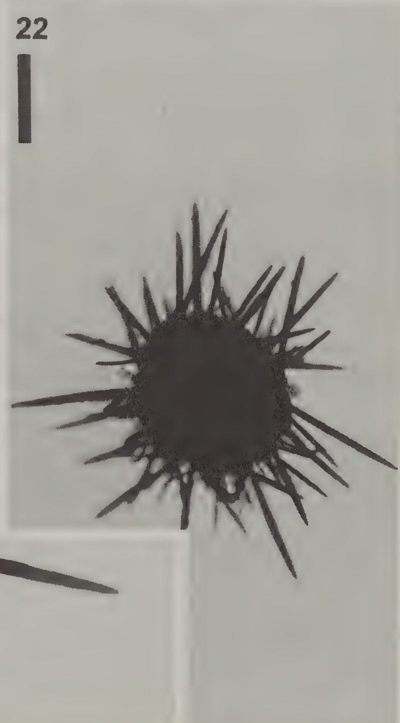

upper half provided with scattered, dark brown, thick-walled, opaque, oval, protruding cells. Perithecia seated on a sparse, pale subiculum; hyphae growing from the base of the perithecia, brown, septate, branched, 5-8.5 µm wide.—Setae spine-like, stiff, upright, acute, thick-walled, dark brown to black, opaque, nonseptate, unbranched or seldom branched near the lower part, to 190 µm long, 6-8 µm wide in the middle and 13-18 µm wide at the bulbose base; the smaller acute setae arising from around the ostiolar region 21-46 µm long and 4-6 µm wide in the middle and 9-11 µm wide at the base. The perithecial apex truncate to broadly rounded, formed of conspicuous clavate to cylindrical cells surrounding the pore; cells thick-walled, dark brown 3-4 µm wide and 7-9 µm high.—Peridium leathery, in surface view *textura angularis*, lateral wall 20-26 µm wide, consisting of two layers. Outer layer of cells thick-walled, brown, polyhedral cells 4-6 µm wide; inner layer of thin-walled, subhyaline, compressed cells.—Ostiolar canal periphysate.—Paraphyses cellular, delicate, early deliquescent.—Asci unitunicate, 8-spored, (68-)71-86(-91) × (11-)12-15(-16) µm, cylindrical to clavate, short stipitate, broadly rounded to truncate above, slightly thickened at the top, with an indistinct, J- apical annulus.—Ascospores ellipsoidal with broadly rounded ends, straight or inequilateral, (13-)14-17(-18) × (4-)5-6.5(-7.5) µm, 4-celled, the two middle cells pale brown to greyish-brown, the two end cells subhyaline to hyaline, not constricted, not thickened at the septa, smooth, 2-seriate with ends partially overlapping in the ascus.

ANAMORPH IN NATURE. Colonies effuse, velvety, pale brown.—Mycelium partly immersed, partly superficial.—Conidiophores macronematous, mononematous, unbranched or branched in the lower parts, cylindrical, pale brown, upright, flexuous in the lower parts, to 200 µm long and 5-6 µm wide in the middle, septate at intervals of 17-30 µm in the lower parts, towards the apex the septation at intervals of 4-8 µm, not constricted or slightly constricted at the septa, apical cells rounded or somewhat inflated, 6-7 µm wide; conidiophores arising from superficial hyphae growing from the base of the perithecial initials and mature perithecia.—Conidiogenous cells polytretic, integrated, intercalary and terminal, 4-6 µm long and 5-6 µm wide, perforated by 1-2(-3) dispersed pores in a single conidiogenous cell, the pore about 1 µm diam.—Conidia obovate or clavate, broadly rounded at the apex, 2(-3)-septate, 15-18 × 8-11µm, not constricted or slightly constricted at the

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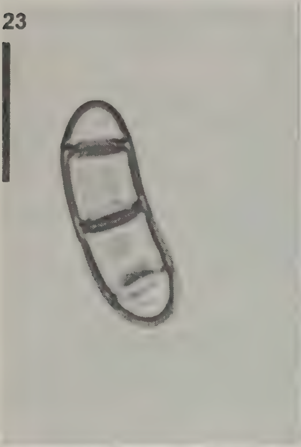
Figs. 23-30. ***Tengiomyces indicus***. 23-25. Ascospores. - 26, 27. Asci with ascospores. - 28-30. Conidia of the ***Spadicoides*** anamorph, from nature. - Figs. 23, 26, 28-30: PC; 27: BF. - Figs. 23-27 from AMH 3871; 28-30 from BPI 622100. - Scale bars: 23-25, 28-30 = 10 µm; 27 = 100 µm.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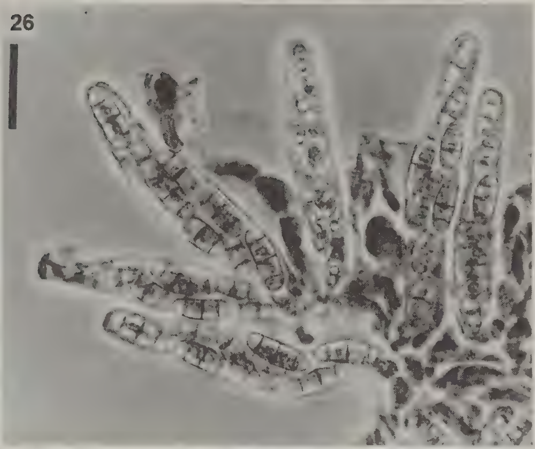
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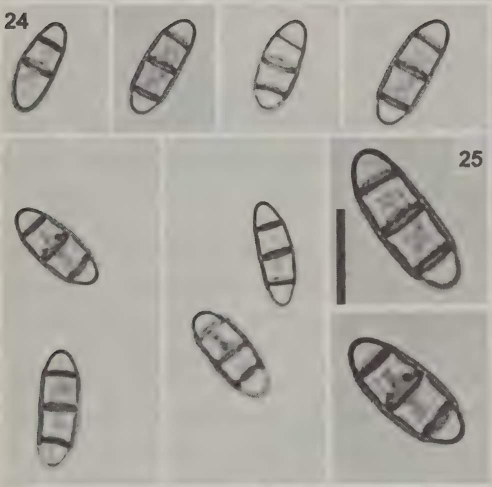

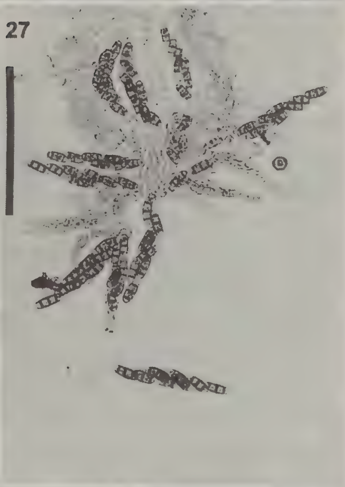
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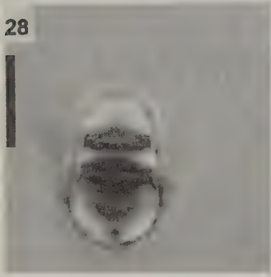
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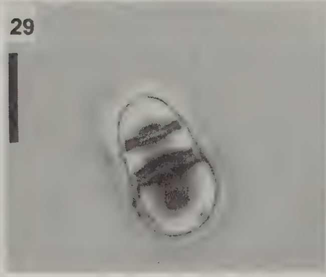
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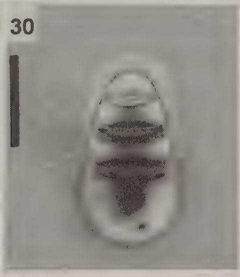
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septa, the septa usually thick and dark with a darker band of colour in the conidium wall, brown, the proximal cell paler than the distal cells or the whole conidium eventualy of the same brown colour, provided with an eccentric pore at the distal end.

MATERIAL EXAMINED. 1) Type material. INDIA. Manjaly, Ekm, Kerala, dead stem of an unidentified dicotyledoneous plant, 2 Oct. 1977, K. I. M. Varghese (AMH 3871 - holotype of *Chaetosphaerella indica*).

2) Additional material. CHINA. Anhwei, Lan-ya-shan, on dead wood of a deciduous tree, 20 June 1933, S. Q. Deng 223 (BPI 622100).

DESCRIPTIONS AND ILLUSTRATIONS. Varghese & Rao (1979: 2, Fig. 2a-d); Teng (1934: 364, Fig. 7).

HABITAT. Lignicolous saprobe on dead stems of an unidentified dicotyledoneous plant and on decaying wood and bark of a deciduous tree.

KNOWN DISTRIBUTION. The fungus is known from Asia only; from China (Teng, 1934) and India (Varghese & Rao, 1979).

DISCUSSION

On the basis of the anatomical characters of perithecia and because of ascal characters and conidiogenesis *Chaetosphaerella* and *Tengiomyces* may be related to *Helminthosphaeria* of the Helminthosphaeriaceae at the familial level. The tretic development of conidium is common among loculoascomycetes (Pleosporaceae, Sivanesan, 1987), but rare among pyrenomycetes where it is restricted to *Helminthosphaeria* and, now to *Chaetosphaerella* and *Tengiomyces*. Ellis (1971b) suggested that the development of conidia is enteroblastic-tretic and the ‚tretoconidia‘ that are solitary or in acropetal chains, are formed ‚by protrusion of the inner wall through one or more channels in the outer wall‘. Cole & Samson (1979) discussed poroconidia and their development in several examples of dematiaceous hyphomycetes, e.g. *Helminthosporium maydis* Nisikado & Miyake, *Ulocladium atrum* Preuss or *Stemphylium botryosum* Wallroth, and concluded that ‚the poroconidium is a special kind of holoblastic propagul.‘ Conidia of the *Oedemium didymum* anamorph of *Chaetosphaerella fusca* are produced in acropetal chains from the apex of the polytretic conidiogenous cells; not more than two chains occur on a conidiogenous cell. The chains are unbranched and are composed of two or more conidia. Conidia of the *Oedemium minus* anamorph of *Chaetosphaerella phaeostroma* develop in a cluster at the apex of the apical swelling of the terminal polytretic conidiogenous cell (Hughes & Hennebert, 1963). Conidia of the *Spadicoides* sp. anamorph of *Tengiomyces indicus* are produced singly from polytretic conidiogenous cells that are terminal and intercalary.

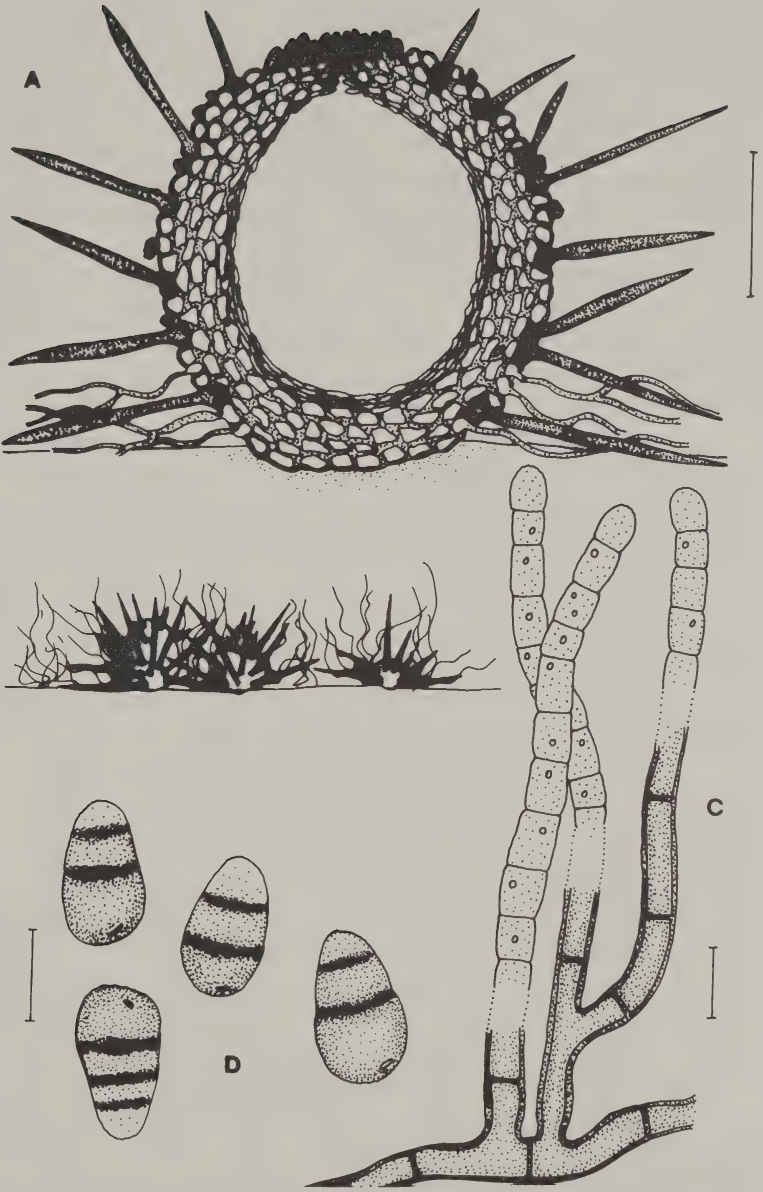

Fig. 31A-D. ***Tengiomyces indicus***. A. Median, longitudinal section of perithecium. - B. Habit sketch of perithecia and perithecia initials accompanied by the anamorph. - C. Conidiophores of the ***Spadicoides*** anamorph, from nature. - D. Conidia of the ***Spadicoides*** anamorph, from nature. - A, B from AMH 3871; C, D from BPI 622100. - Scale bars: A = 100 μm; C, D = 10 μm.

On the basis of the perithecial morphology *Chaetosphaerella* bears some resemblance to the Nitschkiaceae (Fitzp.) Nannf., a highly specialized homogenous group within the Sordariales (Nannfeldt, 1975), but it differs in many other respects, for example in the presence of a definite ostiole, lack of Quellkörper and in ascospore and ascus morphology and anamorphs. *Lasiosphaeriopsis stereocaulicola* Hawksw. of the Nitschkiaceae superficially resembles *Chaetosphaerella* in having 4-celled, versicoloured ascospores and stipitate, obpyriform, tuberculate perithecia but it is distinct in characters of 2-4-spored, long stipitate asci that arise from a basal cushion at the bottom of the perithecial cavity; absence of paraphyses; shape of ascospores; lack of subiculum and an unknown anamorph. *L. stereocaulicola* is a parasite that forms perithecia on the thallus of *Peltigera rufescens* (Hawksworth, 1980).

Chaetosphaerella could be compared with *Crassochaeta* Réblová (Réblová, 1998b) in the character of versicoloured ascospores but it is easily recognized by the obpyriform, tuberculate perithecia that are attached to the substratum by a stout, sterile base; the characteristic structure of the peridium, oblong ascospores and the *Oedemium* and *Veramycina* synanamorphs. Although, *Crassochaeta*, also has 4-celled and versicoloured ascospores, it is distinct by their ellipsoidal shape and septa that are thickened by a dark band; cylindrical, persistent paraphyses; pronounced and refractive apical annulus; subglobose to ovoidal, setose, perithecia and sparse or or dense subiculum of multi-branched setae.

In vertical section of the stalk of *Chaetosphaerella*, divergent vertical rows of cells are observed as is also reported for *Lasiobertia* Sivan., *Lasiophaeriella* Sivan. *Lasiosphaeria ovina* (Pers.: Fr.) Ces. & de Not., *Lasiosphaeriopsis* Hawksw., *Bertia moriformis* (Tode: Fr.) de Not. or *Spinulosphaeria* Sivan. (Sivanesan, 1974, 1975, 1978). Munk pores in the peridial cells, a diagnostic character of all members of the Nitschkiaceae, are also known in several other genera, viz. *Lasiobertia* Sivan. (Sivanesan, 1978), *Lasiosphaeriella* (Sivanesan, 1975) and *Lasiosphaeria* Ces. & de Not., e.g. *Lasiophaeria hirsuta* (Fr.) Ces. & de Not. (= *Lasiosphaeria tuberculata* Carrol & Munk, Carrol & Munk, 1964; Podlahová, 1974).

Tengiomyces indicus cannot be adequately included in any other previously described genus. In the Helminthosphaeriaceae *Tengiomyces* differs from *Helminthosphaeria*, the type genus of the family, in early deliquescing paraphyses and versicoloured, nonporate ascospores. The paraphyses have not been seen in young or mature perithecia, only large, hyaline, delicate cellular fragments 36-43 µm long and 11-14 µm wide have been seen arising among the asci, perhaps remnants of the paraphyses.

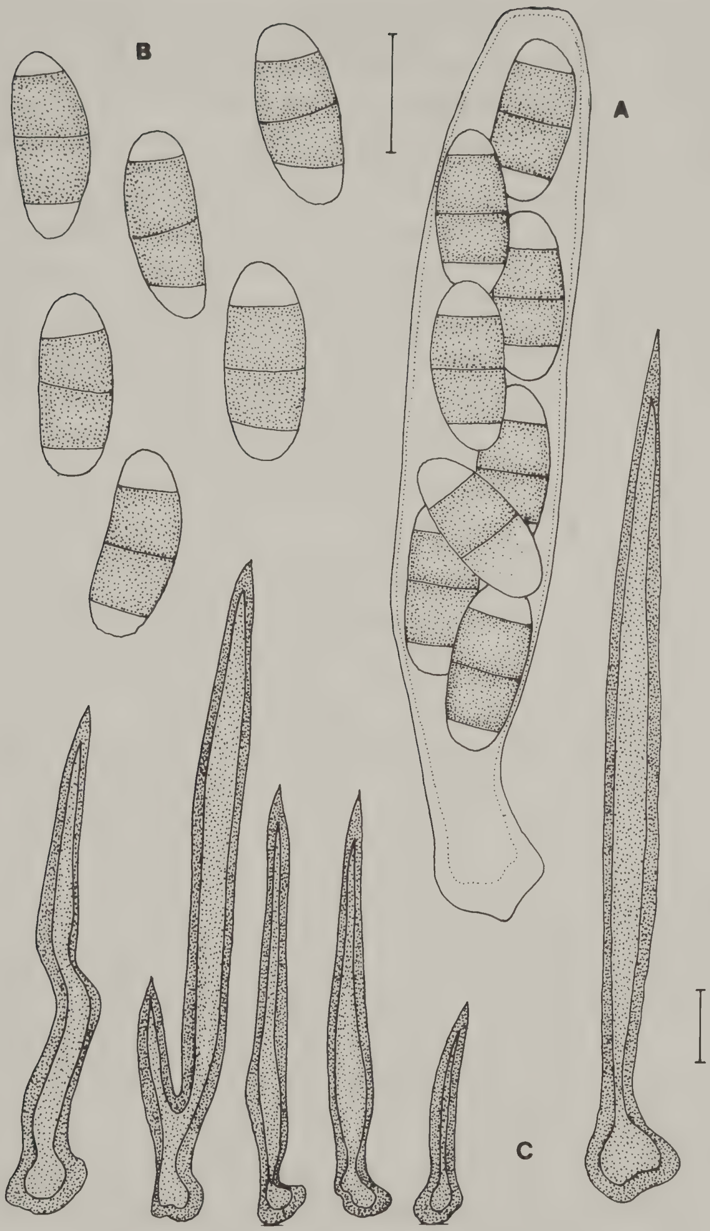

Fig. 32A-C. ***Tengiomyces indicus***. - A. Ascus with ascospores. - B. Ascospores. - C. Perithecial setae. - A-C from AMH 3871. - Scale bars: A-C = 10 μm.

In the form of ascospores *Tengiomyces* resembles *Ascocodinaea* Samuels & al., *Crassochaeta* Réblová and *Chaetosphaerella*. *Ascocodinaea* differs in characters of the peridium; asci with pronounced apical annulus; persistent, cylindrical paraphyses and the anamorph with enteroblastic-phialidic development of conidia, that belong to *Dictyochaeta* Speg. The seta-type observed in *Tengiomyces indicus* is much like that of *Crassochaeta nigrita* (Berk. & Broome) Réblová, in which the setae grow from the perithecia and the similar, but dichotomously or multi-branched setae that arise from the subiculum (Réblová, 1998b). In *Chaetosphaerella phaeostroma*, similar setae growing from the basal part of the perithecium are also present in the subiculum among conidiophores of the anamorph. These rigid, opaque and acute, spine-like setae are also known in some *Coniochaeta* species. *Chaetosphaerella* has, like *Tengiomyces*, a thin and rather indistinct apical annulus and broadly cellular and early dissolving paraphyses, but it is clearly distinguishable by the large, glabrous and obpyriform perithecia with a sterile stipitate base, the morphology of the ascospore and the *Oedemium* and *Veramycina* synanamorphs. *Crassochaeta* is clearly distinct with its much larger, ovoidal to subglobose, distinctly papillate, setose perithecia; ascus with the distinct refractive apical annulus; persistent cylindrical paraphyses; ellipsoidal ascospores that are strongly thickened at the septa by a dark band and their germination by phialides in *C. nigrita*. In addition, the associated anamorph of *Crassochaeta* is morphologically very different from that of *Tengiomyces*.

Tengiomyces indicus is known from two collections only, both made in Asia and it is the only species in the new genus. I have not seen living specimen of this fungus and I am not aware of reports of *Tengiomyces indicus* having been cultured. *Spadicoides* S. Hughes is presumed to be the anamorph of *T. indicus*. The conidiophores and mycelium were found growing from the perithecia initials and mature perithecia as well as from the surface of the substratum in the collection made by Teng in China (Teng, 1934). In the collection made in India (Varghese & Rao, 1979) no associated hyphomycete was found, only the sparse pale brown subiculum was seen growing from the base of the perithecia. The *Spadicoides* sp. associated with the perithecia of *T. indicus* somewhat resembles *Spadicoides obovata* (Cooke & Ellis) S. Hughes in conidial morphology, conidia of *S. obovata* are similar in size (12.5-16 × 6-8.5 µm) and colour but are truncate at the proximal end and lack any visible pore-like structure at the distal end (Goh & Hyde, 1996). No teleomorph of a *Spadicoides* has previously been reported.

Notes on *Chaetosphaeria iquitosensis*

Chaetosphaeria iquitosensis (P. Henn.) Theiss., in Herb. (Not published.)

MATERIAL EXAMINED. PERU, Iquitos, on leaves of *Bactris* sp., April 1903, M. Ule (PRM 656902, W 20227).

The fungus forms black, slightly roughened, globose to obpyriform perithecia on the leaf surface of *Bactris* sp. accompanied by erect, straight, black setae. It superficially recalls *Chaetosphaerella fusca* in the morphology of the ascospores, that are 4-celled. The two middle cells of each ascospores turne brown at maturity, and the two end cells remain subhyaline to pale brown; one of the middle cells is slightly inflated.

The specimen labeled as *Chaetosphaeria iquitosensis* (P. Henn.) Theiss. is preserved in PRM and W but the combination was never published. It is based on *Meliola iquitosensis* P. Henn. (Hennings, Hedwigia 43: 361, 1904). This fungus is *Asteridiella iquitosensis* (P. Henn.) Hansf. of the Meliolaceae G. W. Martin ex Hansf. (Hansford, Sydowia 10: 48, 1957). For full synonymy refer to Hansford (1961).

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STUDIES IN CHAETOSPHAERIA SENSU LATO II. CONIOBREVICOLLA GEN. & SP. NOV.

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ABSTRACT

The new monotypic genus *Coniobrevicolla* is described with the single species *Coniobrevicolla larsenii*, a saprobic fungus occurring on decaying wood of a deciduous tree in the north temperate zone. Based on the characters of peridium anatomy, ascal and hamathecium anatomy the new genus is placed in the Trichosphaeriaceae. *C. larsenii* is described and illustrated and the relationships with *Crassochaeta* of the Trichosphaeriaceae and *Ascocodinaea* of the Chaetosphaeriaceae are discussed. *Chaetosphaeria subcaespitosa* is considered and the relationship with *Coniobrevicolla* is discussed.

Key words. Ascomycotina, Trichosphaeriaceae, wood-inhabiting fungi, systematics.

INTRODUCTION

This is the second paper in a series of publications dealing with twenty-eight species that have 3-7-septate versicoloured ascospores, most of which have been placed in *Chaetosphaeria* Tul. & C. Tul. and fewer in *Chaetosphaerella* E. Müll. & C. Booth and *Lasiosphaeria* Ces. & de Not. Recently, during a revision of *Chaetosphaeria*, a herbarium specimen preserved in C (Denmark) and labelled as *Chaetosphaeria subcaespitosa* P. Larsen was examined. The herbarium specimen does not match the generic concept of *Chaetosphaeria* given by Tulasne & Tulasne (1863). It has glabrous perithecia that collapse by lateral pinching when dry; a wide peridium comprising thick-walled, angular cells; an ascal apex with an inconspicuous apical annulus; filiform, cylindrical paraphyses; 3-septate, versicoloured ascospores with the middle cells becoming brown and the end cells remaining hyaline. No anamorph was associated with the perithecia. *Chaetosphaeria* is distinct in that the perithecia, though also glabrous, are never collabent; the peridium is thin and comprises thin-walled, opaque, brick-like cells; the ascal apex has a pronounced apical annulus; paraphyses are tapered; ascospores are 1-3-septate and generally hyaline. Anamorphs have enteroblastic-phialidic conidiogenesis and are dematiaceous hyphomycetes.

Based on the chracters of the ascospores the herbarium specimen labelled as *Chaetosphaeria subcaespitosa* suggests an affinity with *Ascocodinaea* Samuels & al., a fungicolous genus of the Chaetosphaeriaceae Réblová & al., and with lignicolous *Crassochaeta* Réblová (Réblová, 1998b) of the Trichosphaeriaceae G. Winter but neither proved to be a home for it. These genera are distinct in the anatomy of the peridium, ascal and hamathecium anatomy and the anamorphs. Although the herbarium specimen exhibits some characters typical of *Chaetosphaeria subcaespitosa* described by Larsen (1952) the similarity is only superficial. The fungus labelled as *Chaet. subcaespitosa* could not be placed in any known genus of pyrenomycetous ascomycetes. Therefore, it is described as a new genus *Coniobrevicolla* with the single species *Coniobrevicolla larsenii*. On the basis of the characters of peridium, ascal and hamathecium anatomy the new genus is placed in the Trichosphaeriaceae.

Chaetosphaeria subcaespitosa P. Larsen is considered and its relationship with *Coniobrevicolla* is discussed.

MATERIALS AND METHODS

For materials and methods refer to the first part of this series of publications (Réblová, 1998a).

TAXONOMIC PART

Coniobrevicolla Réblová, gen. nov.

Perithecia superficialia, conica, exsiccata collabescentia, hyphis subiculi sparsis insidentia, pariete ascomatibus 2-stratoso. Ostiolo periphysibus instructo. Paraphyses persistentes. Asci 8-spori, apice annulo haud profundo sed distincto. Ascospores ellipsoideae, 3-septatae, versicolores.

Species typica. Coniobrevicolla larsenii Réblová

Perithecia superficial, conical, papillate, ostiolate, collapsing when dry, at the base with a sparse subiculum.—Peridium consisting of two layers.—Ostiolar canal periphysate.—Paraphyses persistent.—Asci 8-

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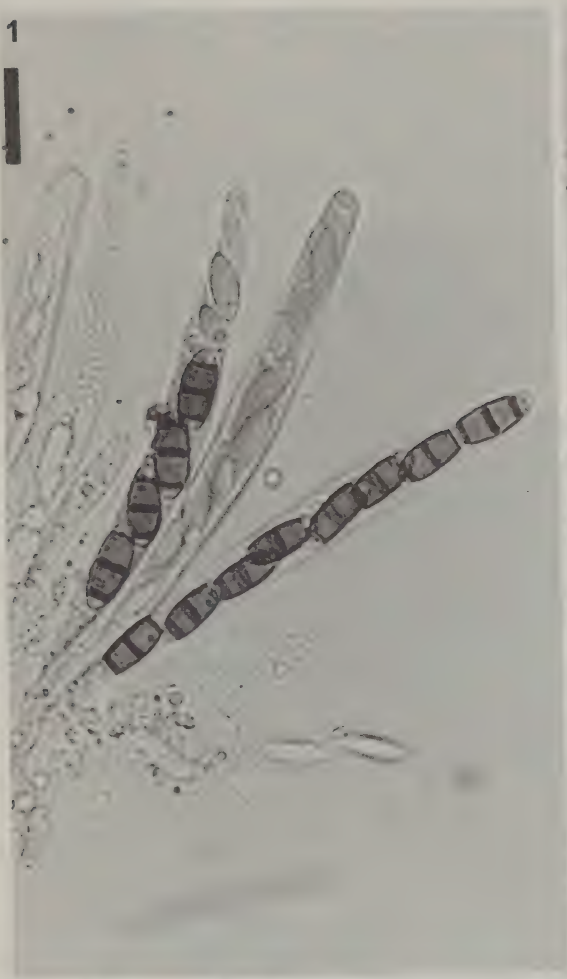
Figs. 1-5. **Conibrevicolla larsenii**. - 1, 2. Asci containing ascospores. - 3 - 5. Ascospores. - Figs. 1-4: BF; 5: PC. - Figs. 1-5 from C. - Scale bars: 1-4 = 20 µm; 5 = 10 µm.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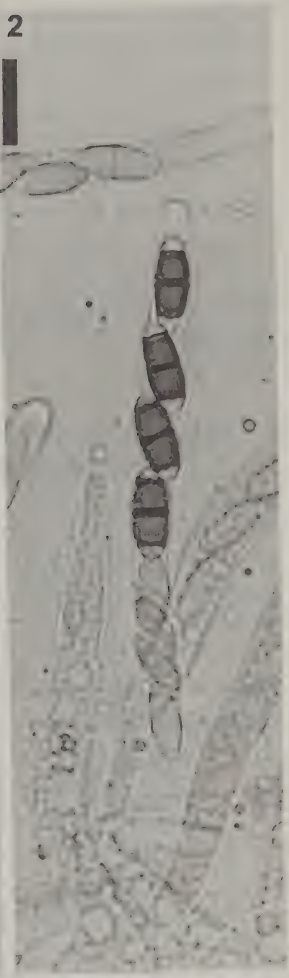
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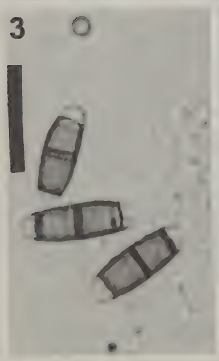
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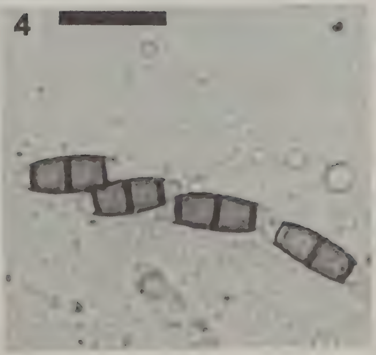
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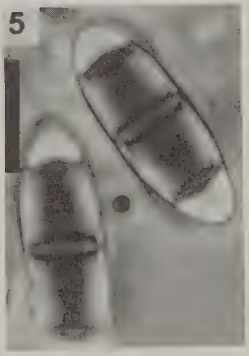
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spored, with an indistinct apical annulus.—Ascospores ellipsoidal, 3-septate, versicolour, the two middle cells becoming brown, the two end cells remaining hyaline.

TYPE SPECIES. *Coniobrevicolla larsenii* Réblová

ANAMORPH. Unknown.

SYSTEMATIC POSITION. Trichosphaeriaceae G. Winter, Trichosphaeriales M.E. Barr

ACCEPTED TAXA. *Coniobrevicolla larsenii* Réblová

ETYMOLOGY. „conio" is derived from the latin *conicus* = conical, refering to the shape of the perithecium; „brevicolla" is derived from the latin *brevicollis* = short neck, refering to the short papillate apex of the perithecium.

Coniobrevicolla larsenii Réblová, sp. nov. Figs. 1-7.

Perithecia superficialia, basi subimmersa, hyphis subiculi sparsis insidentia, conica usque subglobosa, papilla minuta, exsiccata collabescentia 200-250 μm lata, 250-300 μm alta, pariete coriaceo-carbonaceo, extus textura epidermoidea tecta. Pariete ascomatibus 2-stratoso. Ostiolo periphysibus instructo. Asci 8-spori, cylindraceo-clavati, apice annulo haud profundo sed distincto, subincrassato, non amyloideo, (116-)132-147(-156) × 12.5-13.5 μm. Paraphyses persistentes, copiosae. Ascosporae uniseriatae usque oblique uniseriatae, 4-cellulares, versicolores, cellulis duabus centralibus fuscis, apicalibus hyalinis, (19-)20-23(-25) × 7-8.5 μm.

Holotypus. Dania, Sorø Sønderskov, Næstvedvej, in ligno putrido, 18 Oct. 1983, leg. U. Søchting (C).

ANAMORPH. Unknown.

Perithecia superficial with base slightly immersed, scattered or in small groups of 2-3, conical, distinctly papillate, 200-250 μm wide and 250-300 μm high, ostiolate, collapsing by lateral pinching when dry, dark brown to black, glistening, slightly roughened, glabrous, at the base with a sparse subiculum; hyphae brown, septate, branched, 3-4 μm wide.—Peridium leathery to brittle, in surface view comprising cells forming a network (*textura epidermoidea*); lateral wall consisting of two layers. Outer layer 20-28 μm wide, of thick-walled, brown, angular cells that are strongly thick-walled at the surface. Inner layer 3-5 μm wide, of thin-walled, nonpigmented, elongated, compressed cells.—Ostiolar canal periphysate.—Paraphyses persistent, abundant among the asci, filiform, cylindrical, 1.5-2 μm wide, septate, branched, not constricted at the septa.—Asci unitunicate, 8-spored, (116-)132-147(-156) x 12.5-13.5 μm, arising in a broad hymenium, cylindric-

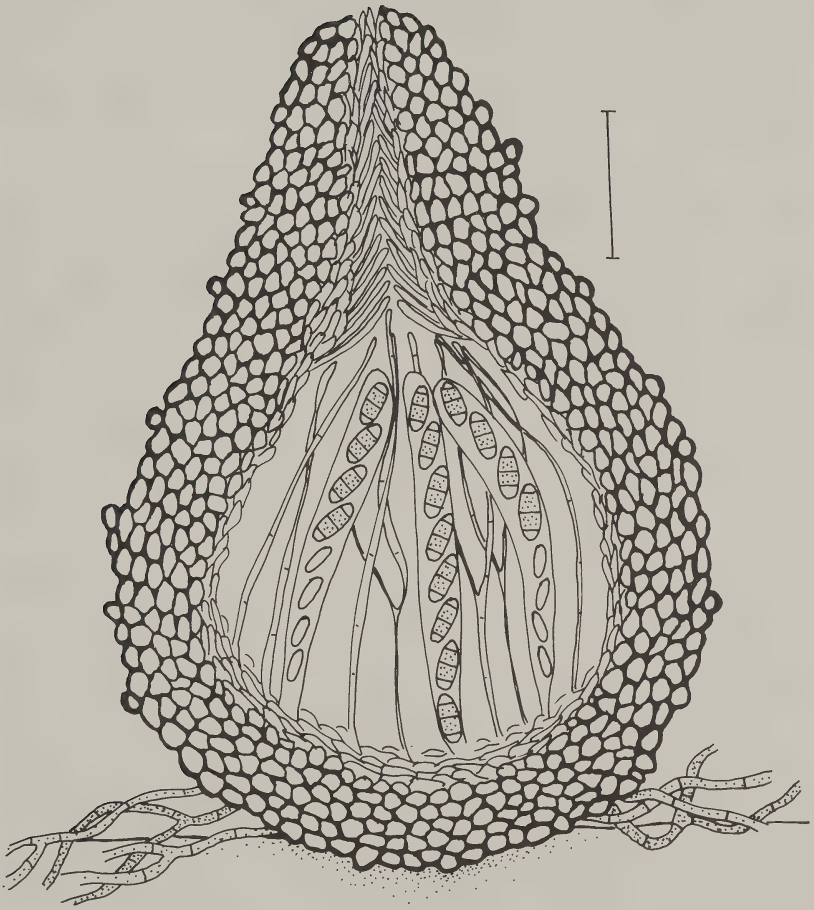


Fig. 6. ***Coniobrevicolla larsenii***. - Median, longitudinal section of the perithecium. Scale bars = 50 µm.

clavate, truncate, apex somewhat thickened with a J-, rather indistinct apical annulus.—Ascospores ellipsoidal with narrowly rounded ends, (19-)20-23(-25) × 7-8.5 µm, 4-celled, the two middle cells becoming brown, the two terminal cells remaining hyaline, slightly constricted at the septa, 1-seriate to obliquely 1-seriate in the ascus.

MATERIAL EXAMINED. DENMARK. Sorø Sønderskov, Næstvedvej, on rotten wood, 18. Oct. 1983, U. Søchting (C, holotype).

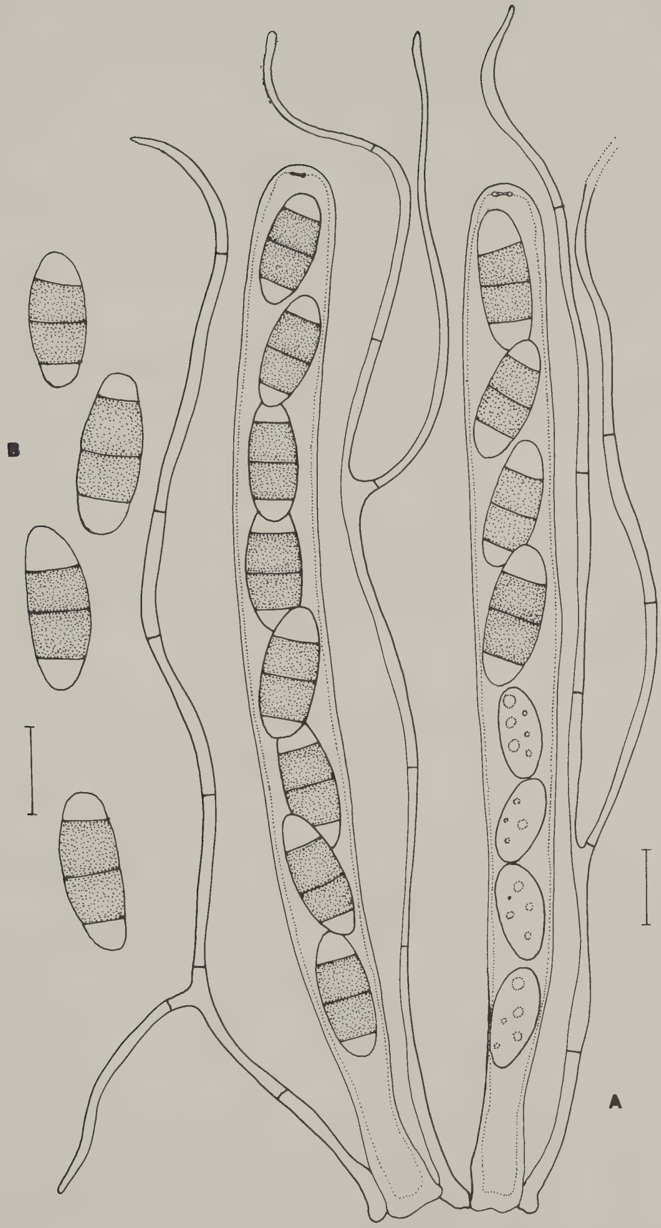
HABITAT. Saprobe on decayed wood of an unidentified deciduous tree.

KNOWN DISTRIBUTION. Europe: Denmark, known only from the type locality.

The type collection of *Chaetosphaeria subcaespitosa* (DENMARK. Jutland, Bramdrup Skov near Kolding, on a decorticated branch of *Sambucus nigra*, March 1921, P. Larsen, C) is apparently lost. The identity of this fungus remains obscure. It was not possible to identify it based on the description and illustration only (Larsen, 1952). The fungus was described and illustrated by Larsen (1952) from a collection made in Denmark on wood of *Sambucus nigra*. The description is taken from Larsen (1952): Ascomata superficial, black, partly immersed in a greyish, felty mass consisting of hyaline hyphae and a granular substance. Asci 115-140 [*pars sporifera*] x 8-10 µm, thick-walled at the top, paraphyses indistinct or absent, ascospores 17-22 x 6-7 µm, ellipsoidal to fusiform, 4-celled, the middle cells becoming brown, the end cells remaining hyaline.

According to the original description the fungus resembles *Coniobrevicolla larsenii*. Larsen (1952) described the appearance of the ascomata, which are immersed in a remarkable light coloured layer. On the basis of the bright hyphal cover Munk (1957) suggested a relationship with *Lasiosphaeria ovina* (Pers.: Fr.) Ces. & de Not.

Although *Coniobrevicolla larsenii* exhibits some characters typical of *Chaet. subcaespitosa*, for which only the original description and illustration remain (Larsen, 1952: 8, Fig. 2), the recently found specimen can hardly be identified as *Chaet. subcaespitosa*. The perithecia of the recent find, although similar to those of Larsen´s fungus in size, colour and the absence of setae, lack the greyish hyphal layer that was described by Larsen (1952) as covering the ascomata of *Chaet. subcaespitosa*. The covering could be a third, light coloured, hyphal layer over the perithecia, a typical character of some species of the Lasiosphaeriaceae Nannf., e.g. *Bombardia macrocarpa* Carroll & Munk, *Lasiosphaeria chrysentera* Carroll & Munk, *Lasiosphaeria ovina* (Pers.: Fr.) Ces. & de Not. or of the Chaetosphaeriaceae Réblová & al., e.g. *Melanochaeta* E. Müll., Harr & Sulmont (Réblová & al., 1998). In



Figs. 7A-B. ***Coniobrevicolla larsenii***. - A. Asci with ascospores and paraphyses. - B. Ascospores. - A, B from C. - Scale bars: A, B = 10 µm.

these fungi the light coloured layer often disappears with increasing age, leaving the perithecia black and glabrous. This seems to be the case with *Chaet. subcaespitosa* as well. Unfortunately, the type collection of *Chaet. subcaespitosa* was not found in C and I cannot say with certainty what the hyphal layer described by Larsen was. Both collections, the recent and the original, originate from Denmark, but comparision *Coniobrevicolla larsenii* with description of the type specimen of *Chaetosphaeria subcaespitosa* indicates that the latter differs in the anatomy of the perithecial peridium. Larsen neither described and illustrated paraphyses, but they were copiously produced and persistent in *Coniobrevicolla larsenii.*

DISCUSSION

In the characters of the ascospores *Coniobrevicolla* resembles *Ascocodinaea* of the Chaetosphaeriaceae and *Crassochaeta* of the Trichosphaeriaceae. *Crassochaeta* (Réblová, 1998b) would seem to be a possible home for *Coniobrevicolla* in having 3-septate, versicoloured ascospores but both species included in *Crassochaeta*, viz. *Crass. fusispora* and *Crass. nigrita*, are distinct in having setose, roughened perithecia; a pronounced apical annulus; wide (8-11 µm); paraphyses that are cylindrical, slightly tapered and at the septa constricted. Subiculum that is present in *Crass. nigrita* is dense, black of dichotomously or multibranched, sterile, spine-like setae.

Two species of *Ascocodinaea* are described (Samuels & al., 1997) as fungicolous saprobes that form setose perithecia on the old hymenium of members of the Aphyllophorales. *Ascocodinaea* differs from *Conibrevicolla* most conspicuously in that the perithecial peridium in *Ascocodinaea* is in translucent brown, not melanized, and comprises thin-walled, brick-like cells. The perithecial apex of *A. stereicola* is formed of enlarged cells arranged in rows. The apex of *A. polyporicola* is not anatomically distinct from the wall below but the vertical section of the upper part of the perithecium shows the hyphal character of the cells. The anamorph belongs to *Dictyochaeta* Speg. (as *Codinaea*, Samuels & al., 1997). The present author accepts the name *Dictyochaeta* for the anamorph of *Ascocodinaea* as the correct name for the taxon with *Codinaea* Maire as its synonym, in accordance with Gamundi & al. (1977), Kirk (1982) and Holubová-Jechová (1984). The type species of *Dictyochaeta*, *D. fuegiana* Speg., was recollected and redescribed by Gamundi & al. (1977) and *Codinaea* Maire was listed as the synonym. At present, about 70 species are described in both *Codinaea* and *Dictyochaeta.* Kuthubutheen & Nawawi (1991) considered *Dictyochaeta* and *Codinaea* as distinct taxa and listed the species accommodated in *Codinaea* and *Dictyochaeta* along with a key. The authors considered the anamorphic conglomeration *Dictyochaeta/Codinaea* and its teleomorphs (*Ascocodinaea*, *Chaetosphaeria*, *Striatosphaeria* Samuels & E. Müll.) and distributed species of both *Dictyochaeta* and *Codinaea* among several groups according to morphological characters of conidia, conidiogenous cells and setae, without considering their teleomorph-anamorph connections.

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TWO NEW SPECIES OF *CALICIUM* FROM EUROPE

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ABSTRACT

The new European species *C. montanum* and *Calicium pinastri* (Caliciaceae) are described. *C. montanum* occurs on wood and ranges from the Alps to the Mediterranean. *C. pinastri* grows on bark and is known from the Czech Republic, Germany and Finland. A key to the seventeen European species known is supplied.

INTRODUCTION

Calicium is a well-known genus of "pin-lichens", and seventeen species are now known to occur in Europe. Keys to the species have been provided in several papers (Poelt 1969, Clauzade & Roux 1985), but they are very incomplete.

Most species occur on bark or lignum, mainly on old trees in old-growth forests or parklands. Several of them are red-listed in different countries and they have frequently been used together with other pin-lichens in biomonitoring of lichen and organism diversity, forest stand structure and continuity, and ecology (Holien 1996, Hyvärinen et al. 1992, Rikkinen 1995, Tibell 1992).

Calicium species usually have rather long-stalked apothecia, although stalk length like many other morphological features exhibit strong variation, and occasional short-stalked morphotypes do occur in most species. Of the European species so far described, only *C. corynellum* (Ach.) Ach. and *C. diploellum* Nyl. consistently have very short-stalked apothecia, whereas in *C. adspersum* Pers. the stalks are sometimes rather long. Like these species, *C. montanum* also has with short-stalked apothecia and pruinose capitula. *C. pinastri* is a minute species somewhat similar to *C. glaucellum* (Ach.) Ach. and *C. parvum* Tibell

MATERIAL AND METHODS

For SEM, dry material was mounted on specimen stubs and coated with gold. For transmission electron microscopy, small pieces of material were fixed in 2.5% glutaraldehyde buffered with sodium cacodylate at pH 7.2, post-fixed in 2% osmium tetroxide at 4°C for two hours, embedded in Epon and sectioned and double-stained with lead citrate and uranyl acetate. Estimates of the apothecium height, capitulum width, stalk width, ascus length, ascus width, spore length and spore width are given in the following way: a-b, or for combinations of length and width a-b x c-d, where a (and c) represent the arithmetic mean minus 1 standard deviation and b (and d) the arithmetic mean plus 1 standard deviation. The arithmetic mean (X) and the number of observations (n) are also supplied.

RESULTS

Calicium montanum Tibell, sp. nov.

Figs 1-2.

Thallus bene evolutus, irregulariter granulosus ad verrucosum, pallidocinereus ad viridulum. Apothecia breve stipitata, pruina distincta alba in externa pagina excipuli, 0.3-0.4 mm alta. Capitulum obconicum ad lenticulare, 0.19-0-31 mm diametro. Stipes brevis latusque, 0.11-0.14 mm diametro. Asci ubi maturi cylindrici, sporis uniseriate ordinatis, 39-41 x 5-6 µm. Sporae ubi juvenes laeves, ubi maturae ornamento rimarum irregularium, late ellipsoidales, 9.5-11.5 x 5-6 µm.

Type: Germany. Bavaria, Ammergauer Alpen, between Neuschwanstein and Bleckenau, 1971 Tibell 4548 (holotype, UPS).

Thallus well-developed, irregularly granular to verrucose, pale grey to greenish. Apothecia short-stalked, 0.32-0.41 mm high (X=0.36 mm, s=0.043 mm, n=30), with a distinct white pruina on the outer surface of the excipulum. All parts of the apothecia I- (watermount of sections or squash preparations).

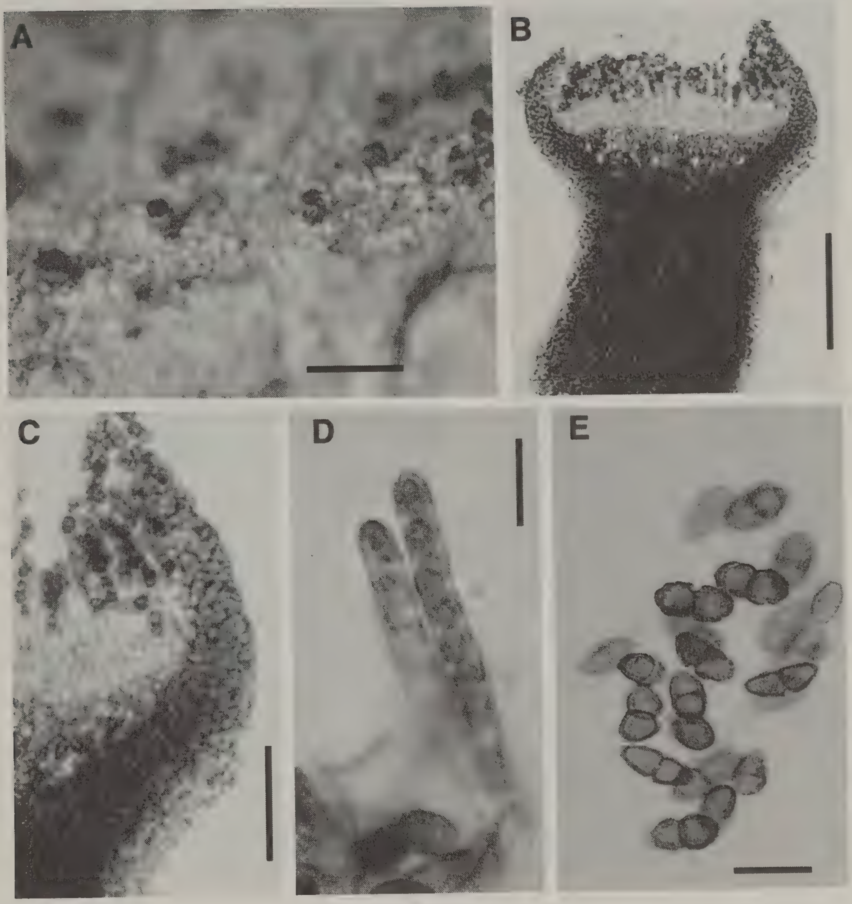

Fig. 1. *Calicium montanum*. A. Mature apothecia on granular thallus. Bar = 0.5 mm. B. Longitudinal section of mature apothecium. Bar = 100 µm. C. Radial section of excipulum. Bar = 50 µm. D. Mature asci. Bar = 10 µm. E. Mature spores. Bar = 10 µm. - A: Germany, Oberhollenzer 2696a (hb. Oberhollenzer). B-C: Spain, Burgaz 1007 (hb. Burgaz). D-E: Germany, Oberhollenzer 2459a (hb. Oberhollenzer).

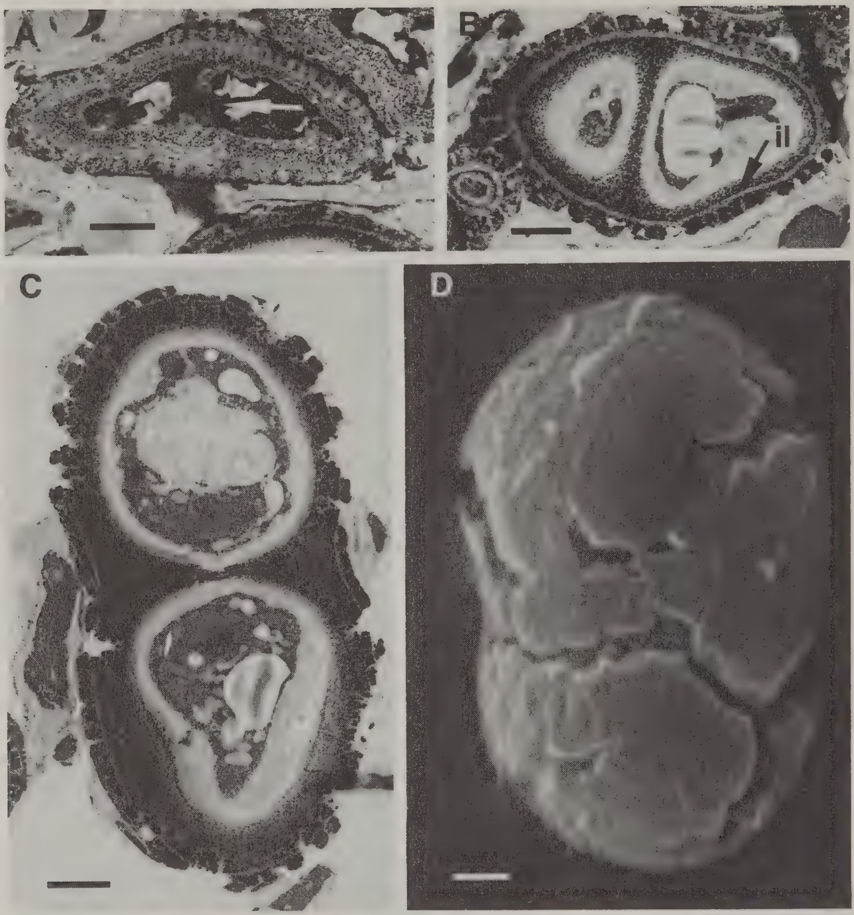

Fig. 2. *Calicium montanum.* A. Longitudinal section of young spore still contained in its ascus with young septum indicated by an arrow. Bar = 1 µm. B. Longitudinal section of semi-mature spore released from its ascus. Intermediary layer (il) clearly visible separating the proper wall from the epispore. Bar = 1 µm. C. Longitudinal section of mature spore with irregularly ruptured perispore over a strongly thickened and pigmented proper wall. Bar = 1 µm. D. Mature spore. Bar = 1 µm. - A-D: Portugal, Pargue Natural de Monteshinho, 1995 v. d. Boom (hb. v. d. Boom).

Capitulum obconical to lenticular, 0.19-0.31 mm diam. (X=0.25 mm, s=0.058 mm, n=30). Excipulum formed by dark brown to aeruginose, almost isodiametric sclerotized cells. Hypothecium dark brown, with a flat or slightly convex upper surface. Stalk short and wide, 0.11-0.14 mm diam. (X=0.12 mm, s=0.025 mm, n=30), consisting of dark brown, irregularly interwoven and strongly sclerotized hyphae. Surface layer of the stalk paler and forming a distinct, hyaline, I- coat. Asci when mature cylindrical, with uniseriately arranged spores, 38.6-41.4 x 4.7-5.9 µm (X_l=40.0 µm, s=1.4 µm, n=28; X_w=5.3 µm, s=0.6 µm, n=zx). Spores when young smooth, when mature with an ornamentation of irregular cracks, broadly ellipsoidal, 9.6-11.4 x 4.8-6.1 µm (X_l=10.8 µm, s=0.9 µm, n=33; X_w=5.5 µm, s=0.7 µm, n=30).

Spore ontogeny. In TEM the semi-mature spores have a very distinctive, electron-lucent intermediate layer separating the perispore from the proper wall (Fig. 2B). The perispore is 0.25-0.35 µm thick, and consists of radially arranged electron-dense elements separated by narrow, or sometimes irregularly widened, electron-lucent furrows. The proper wall is 0.20-0.25 µm thick and provided with numerous minute pigment granules. Endospore electron-lucent, often irregular. In younger spores (Fig. 2A), the wall layers are visible, but less pigmented and less clearly delimited than in semi-mature spores. Here the septum initial is still thin and winding. In the fully mature spores (Fig. 2C) the electron-dense parts of the perispore are more or less fragmented to form an irregular ornamentation resting on the intermediate layer. Occasionally, however, the cracks of the perispore continue into the proper wall, which is now 0.45-0.65 µm thick and densely pigmented by fused pigment granules. Both the proper wall and the endospore contribute to the septum, which has an electron-lucent mid-lamella. In SEM (Fig. 2D) the spores are seen to have a rather smooth, but irregularly ruptured surface.

Chemistry. Thallus K-, C-, KC-, Pd-; containing divaricatic and 2-O-methyldivaricatic acid (originally identified by Dr. J. Elix, Canberra, pers. comm.).

Habitat. On wood of *Castanea* in ancient woodlands and by fields in the Mediterranean area, in open situations on lignum of *Juniperus oxycedrus* and *Quercus pyrenaica* in Spain, and on wood of *Picea abies* and *Pinus mugo* in forests in the Alps. Alt. 700-900 m.

Distribution. Known from rather few collections in Italy, Spain, Portugal and Germany. The species possibly has a slightly oceanic distribution. Probably overlooked

Note. Characterized by its often very thick, verrucose and slightly granular thallus, the sturdy, short-stalked, pruinose apothecia, and the medium-sized spores which are smooth as young and when mature have a coarse ornamentation of irregular cracks. It is sometimes similar to *C. quercinum*, which however differs in having rather slender apothecia, a K+ yellow-red, Pd+ yellow thallus containing norstictic acid, and in having smaller spores with an ornamentation of spirally arranged ridges.

Specimens examined. *Germany*, holotype; Allgäu, Wildpoldsried, 1988 Oberhollenzer 2459a (hb. Oberhollenzer); Unterschwarzenberg, 1989 Oberhollenzer 2696a (hb. Oberhollenzer). *Italy*. Toscana, Lucca prov., N of Bagni di Lucca, 1985 Coppins 12124 (E), James & Rose. *Portugal*. Trás os Montes, NE of Villa Real, N of road to Murca, 1995 v. d. Boom (hb. v. d. Boom); Pargue Natural de Monteshinho, 10 km W of Braganca, 1995 v. d. Boom (hb. v. d. Boom). *Spain*. Toledo, Montes de Toledo, Aragón, 1995 Burgaz (hb. Burgaz).

Calicium pinastri Tibell, sp. nov.

Figs 3-4.

Thallus tenuissimus, cinereus vel leviter viridulus, vel immersus. Apothecia parva, 0.3-0.4 mm alta. Capitulum obconicum ad lenticulare, 0.14-0.20 mm diametro. Stipes saepe brevis, niger, nitidus, epruinosus, 0.09-0.15 mm diametro. Asci ubi maturi cylindrici, sporis uniseriate ordinatis, 30-35 x 3.5-5.5 µm. Sporae ubi juvenes laeves, ubi maturae ornamento rimarum irregularium, late ellipsoidales, 9.5-13.5 x 5-6.5 µm.

Type: Finland, Ostrobottnia media, Lappajärvi, Isaks park, 6.IV.1907 Backman (holotype, H).

Thallus very thin, grey or with a greenish tinge, or immersed. Apothecia small, 0.33-0.40 mm (X=0.37 mm, sd=0.036 mm, n=40) high, epruinose. All parts of the apothecia I- (watermount of sections or squash preparations). Capitulum obconical to lenticular, 0.14-0.20 mm (X=0.17 mm, sd=0.032 mm, n=40) diam. Excipulum formed by dark brown, elongated to almost isodiametric strongly sclerotized cells, which are paler and anticlinally arranged close to the outer surface of the excipulum. In the innermost part of the excipulum the cells are elongated and periclinally arranged. Hypothecium dark brown, with a flat or slightly convex upper surface. Stalk often rather short, shining black, epruinose, 0.09-0.15 mm (X=0.12 mm, sd=0.027 mm, n=40) diam., consisting of dark brown, irregularly interwoven, and strongly sclerotized hyphae. Surface layer of the stalk paler and forming a more or less distinct, hyaline, I- coat. Asci when mature cylindrical, with uniseriately arranged spores, 30-35 x 3.7-5.4 µm (X_l=32.4 µm, sd= 2.72 µm, n=40; X_w=4.6 µm, sd= 0.87 µm, n=40). Spores when young smooth, when mature with an ornamentation of irregular cracks, broadly ellipsoidal, 9.5-13.5 x 5.1-6.4 µm (X_l=11.4 µm, sd= 2.08 µm, n=40; X_w=5.7 µm, sd= 0.64 µm, n=40).

Spore ontogeny. In TEM the semi-mature spores have a distinctive, electron-lucent intermediate layer separating the perispore from the proper wall (Fig.4A). The perispore is 0.20-0.30 µm thick, and contains scattered, minute pigment granules. The proper wall is not distinct from the endospore. In more mature spores (Fig. 4B) the intermediary layer is even more distinct, and the perispore still homogenous and strongly pigmented. The proper wall is also very distinctive and heavily pigmented in a weak, concentric pattern. The outer part of the endospore contains pigment granules. In the fully mature spores (Fig. 4C) the perispore is still homogenous and strongly pigmented, but with irregular fissures, and rests on the intermediary layer. Both the proper wall and the endospore contribute to the septum, which has an electron-lucent mid-lamella. In SEM (Fig. 4D) the spores are seen to have a minute and irregularly verrucose surface with occasional large cracks.

Chemistry. Thallus K-, C-, KC-, Pd-; containing minute amounts of several not identified compounds.

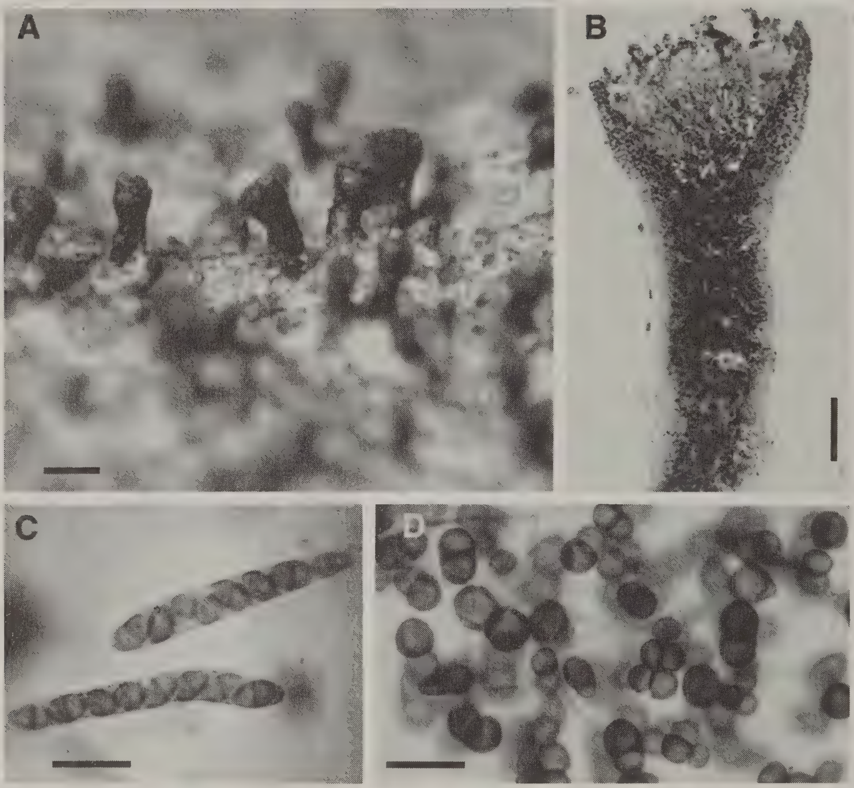

Fig. 3. *Calicium pinastri* A. Mature apothecia. Bar = 0.2 mm. B. Longitudinal section of mature apothecium. Bar = 50 µm. C. Mature asci. Bar = 10 µm. D. Mature spores. Bar = 10 µm. - A-C: Finland, Ostrobottnia media, Lappajärvi, 1907 Backman (H). D: Bohemia, Sumava Mts., 1996 Palice (hb. Palice).

Habitat. On easily flaking bark of trunks of *Pinus sylvestris*, occasionally on bark of *Picea abies*. From low altitudes in Finland to 900-1000 m in Germany.

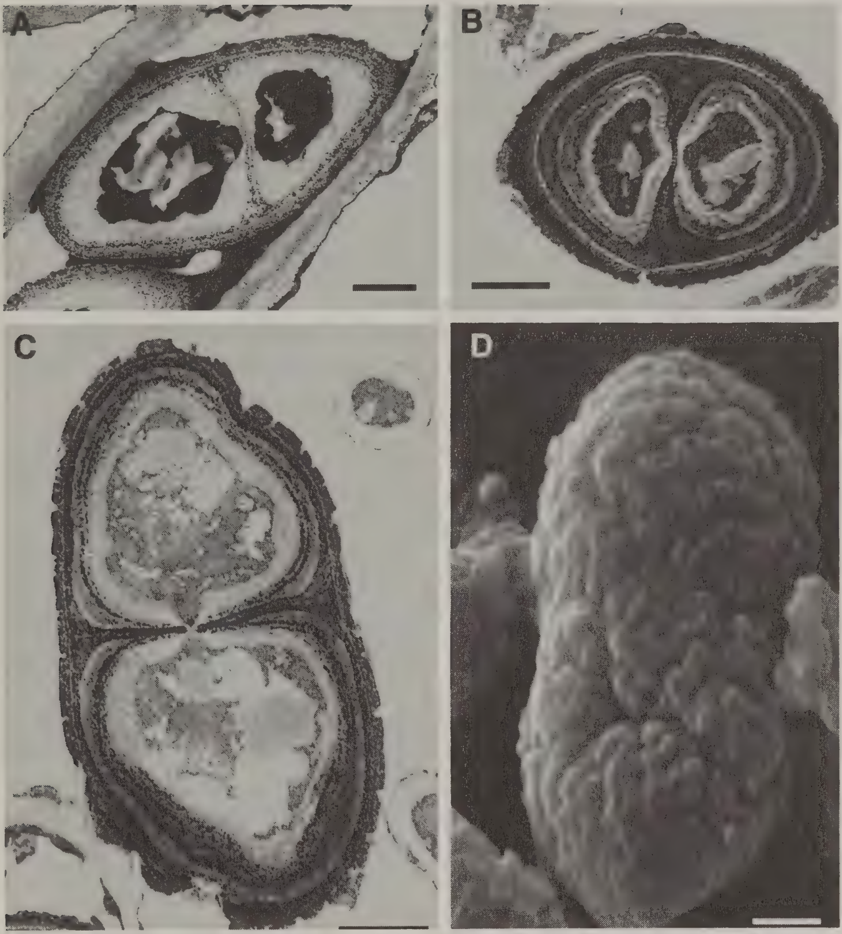

Fig. 4. *Calicium pinastri.* A. Young spore still contained in its ascus. Bar = 1 µm. B. Longitudinal section of semi-mature spore released from its ascus. Intrermediary layer clearly visible separating the proper wall from the epispore. Bar = 1 µm. C. Longitudinal section of mature spore with irregularly ruptured perispore. Bar = 1 µm. D. Mature spore. Bar = 1 µm. - A-D: Finland, Jyväskylä, 1912 Linkola (H).

Distribution. Rather widely distributed in Finland, rare in Bohemia and Germany or possibly overlooked.

Note. Characterized by its thin thallus, minute and often rather short-stalked, epruinose apothecia, the medium-sized spores which are smooth as young and when mature have an ornamentation of irregular cracks. It is quite similar to some specimens of *C. glaucellum.* with small apothecia, but *C. glaucellum* differs in having larger apothecia, in usually having a distinct rim of white pruina at the edge of the excipulum and by containing sekikaic acid as major secondary substance. *C. parvum,* which also usually occurs on bark of *P. sylvestris,* differs by having clavate rather than cylindrical asci, and containing diffractaic acid as major secondary substance. *C. abietinum* differs in usually having brownish stalks and larger spores with an ornamentation of minute warts.

Specimens examined. *Czech Republic.* Bohemia, Sumava Mts., Volary, near Hucina 0.6 km ESE of Cerny Kríz, 48°52'N, 13°52'E, 1996, 1998 Palice (hb. Palice). *Finland.* Nylandia, Sipoo, Söderkulla, 1910 Linkola (H). Ostrobottnia borealis, Muhos, 1923 Räsänen (H). Ostrobottnia media, holotype; Haapajärvi, 1935 Koskinen (H). Savonia borealis, Kuopio, Mustalahti, 1909 Linkola (H); Harjula, 1907 Linkola (H). Tavastia australis, Hollola, Nygård, 1882 Norrlin (H); Lammi, Evo, Mustajärvi, 1909 Backman (H); Jämsä, Pukinvuori, 1935 Koskinen (H). Tavastia borealis, Jyväskylä, Hirvenpelto, 1912 Linkola (H); Jyväskylä, Ylistö, 1912 Linkola (H). "Savolaxia", 1852 E. Nylander (H). *Germany.* Bayern, Rottach-Egern am Tegenersee, NW slope of Mt. Wallberg, 1976 Christiansen (UPS).

KEY TO THE EUROPEAN SPECIES OF CALICIUM.

1 Apothecia with moderate to long stalks, stalks at least twice as tall as the diameter of the capitulum 2

- Apothecia short-stalked or nearly sessile 18

2 Stalk and excipulum in section or squash preparation I-, or only a hyaline surface layer of the stalk slightly I+ reddish blue 3

- Stalk and excipulum in section or squash preparation I+ dark blue to blackish 17

3 Apothecia with a yellow pruina on the mazaedium or the lower side of the capitulum..........4
- Apothecia without yellow pruina..........6

4 Lower side of capitulum with a brown pruina.......... *C. chlorosporum* F. Wilson
- Lower side of capitulum with or without a yellow pruina, not brown..........5
5 Thallus immersed, Pd-. Asci cylindrical.......... *C. trabinellum* (Ach.) Ach.
- Thallus verrucose, Pd+ yellow to red. Asci clavate.......... *C. adspersum* Pers.
6 Lower side of capitulum with a brown pruina..........7
- Lower side of capitulum with or without a white pruina, not brown..........9
7. Asci clavate, thallus green.......... *C. viride* Pers.
- Asci cylindrical, thallus immersed or pale yellowish..........8
8 Thallus immersed or seen as a grey stain of the substrate, UV-, mazaedium never with a yellow pruina, spores 8-11 x 4-5 µm.......... *C. salicinum* Pers.
- Thallus thin, pale yellowish, UV+ yellow or dull, mazaedium often with a yellow pruina, spores 10.5-12.5 x 5-6 µm.......... *C. chlorosporum* F. Wilson
9 Mature asci clavate..........10
- Mature asci cylindrical..........11
10 Thallus grey, thin, areolate, slightly glossy, spores 8-11 x 3.5-5 µm, conidiomata frequent.......... *C. parvum* Tibell
- Thallus green, usually thick, dull, spores 13-14 x 6-7 µm, conidiomata rare.......... *C. viride* Pers.
11 Apothecia usually with a pruina at least along the edge of the excipulum..........12
- Apothecia without pruina..........14
12 Thallus immersed, K-.......... *C. glaucellum* Ach.
- Thallus distinct, grey or pale yellowish, K+ yellow to red..........13
13 Thallus thin, verrucose, smooth, C+ orange, spores without spirally arranged ridges.......... *C. hyperelloides* Nyl.
- Thallus thick, grey, verrucose, with granular surface, C-, spores with spirally arranged ridges.......... *C. quercinum* Pers.
14 Thallus C+ orange.......... *C. hyperelloides* Nyl.
- Thallus C-..........15

15 Stalk often with brownish tinge, mature spores with an ornamentation of minute verrucae, apothecia 6-11 times as high as the diameter of the stalk .... *C. abietinum* Pers.

- Stalk shining black, mature spores with a coarsely areolate ornamentation, apothecia 2-4 or 10-14 times as high as the diameter of the stalk 16

16 Apothecia 0.3-0.4 mm high, 2-4 times as high as the diameter of the stalk, spores 9.5-13.5 x 5-6.5 µm .... *C. pinastri* Tibell

- Apothecia 0.7-1.3 mm high, 9-17 times as high as the diameter of the stalk, spores 11-14 x 6-8 µm .... *C. denigratum* (Vain.) Tibell

17 Stalk pale olivaceous to grey, spores with distinctive ornamentation of spirally arranged ridges .... *C. adaequatum* Nyl.

- Stalk black or very dark reddish brown, spores with a minute, irregular ornamentation ... *C. lenticulare* Ach.

18 Apothecia epruinose or with a greyish pruina 19

- Apothecia with a yellow pruina 20

19 Thallus green to yellowish green, asci clavate, on rocks .... *C. corynellum* (Ach.) Ach.

- Thallus grey to green, asci cylindrical, on lignum .... *C. montanum* Tibell

20 Apothecia 0.1-0.2 mm high, thallus poorly developed .... *C. diploellum* Nyl.

- Apothecia 0.8-1.4 mm high, thallus verrucose to irregular, grey .... *C. adspersum* Pers.

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A BLUE ASCOSPORE COLOUR VARIANT OF EMERICELLA NIDULANS

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ABSTRACT

An *Emericella nidulans* with blue ascospores was isolated during a monitoring of airborne fungal spores in a hospital environment. Morphological studies as well as comparative physiological tests concerning the substrate utilisation showed no significant differences between this isolate and typical *E. nidulans*. The constantly blue pigment of the ascospores alone does not justify to constitute a valid basis for a taxonomic distinction. A key to the 5 known taxa of *Emericella* with blue ascospores is appended.

Key words: *Emericella nidulans, Aspergillus,* ascospores, substrate utilisation.

An *Emericella* isolate belonging to the section *Nidulantes* was cultured during a monitoring of airborne fungal spores in a hospital environment. On close study it resembles *E. nidulans* (Eidam) Vuill., differing from this taxon by the constantly blue-violet to blue-greyish colour of the ascospores.

Raper & Fennell (1965, p. 525) mention that 'Apirion (1963) obtained mutants of *A. nidulans* with blue ascospores by nitrous acid or ultraviolet light'. Apirion treated conidial suspensions of *E. nidulans* with UV and HNO_2; thus, he obtained two types of mutants with blue and white ascospores respectively. The dark blue colour of the blue mutants 'slowly changed to red over a period of a month or more.' In contrast to the description given by Apirion, the blue-violet ascospores of our *Emericella* isolate do not change their pigment over a period of more than 24 months, neither in one and the same culture, nor after several passages. Therefore we describe this interesting blue-spored variant of *E. nidulans*.

Emericella nidulans (Eidam) Vuill.

Material: isolated on DG18 from the air of the University hospital Innsbruck, Austria, 1996-06-18; IB 97/795: a dried culture on MEA.

Cultures deposited: CBS Baarn, The Netherlands, CBS Nr. 100.522, and in the fungal collection of 'Food Science Australia', P.O. Box 52, North Ryde 2113, Australia.

Measurements are given as (minimum) mean value ± standard deviation (maximum), sample size: n = 10-60. $Q_{l/w}$ = quotient length/width, V = volume. M = Colour notations according to Kornerup & Wanscher (1978).

Colonies on CYA (Klich & Pitt, 1988) attaining a diameter of 5-7.5 cm at 25°C in 7 days, plane or somewhat furrowed, producing numerous grass-green (M 30: E 5-7), hemispherical heads in the peripheral colony area, toward the centre increasing numbers of yellowish white clusters of irregular ovoid to ellipsoidal thick-walled Hülle cells; centre with yellowish white (M 4: A2) aerial mycelium and only few conidiophores. Reverse and agar reddish orange (M 7: A-B, 6-7). Exudate clear to pinkish white (M 7: A2-3). Slight mushroom-like odour, not distinctive. Soluble pigment pink.

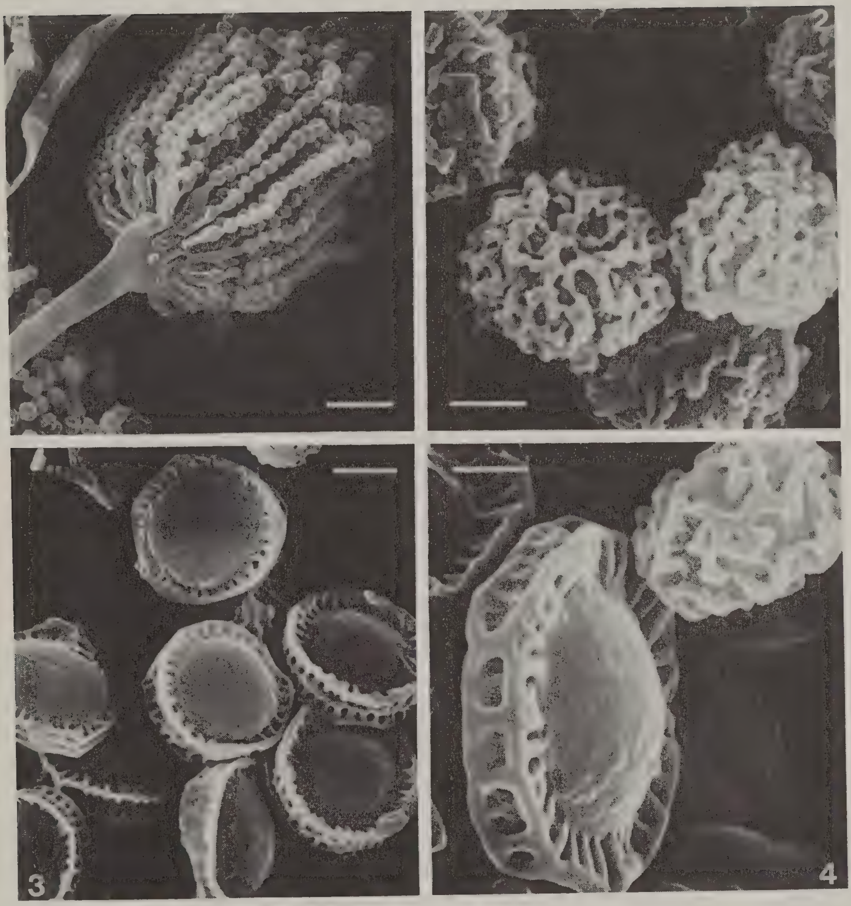
Conidial heads grass-green to greyish green (M 30: E 5-7), generally columnar, but sometimes loosely radiate in age, (47-) 50 ± 6 (-62) µm (n = 20) diameter. Conidiophores straight to slightly sinuous, mostly 90-150 µm in length, by 4.5 to 6.5 µm below the vesicle, basal part 2.5-4.5 µm diameter, wall approximately uniform (0.5-0.8 µm) in width, smooth when young, with amorphous deposits of pigment in age, tan to brown, septate. Vesicles slightly elongate, (8.5-) 11.3 ± 1.1 (-11.8) µm (n = 11), the upper half covered to two thirds by metulae, the lower part sterile. Metulae (5.2-) 5.7 ± 0.4 (-6.4) µm (n = 6). Phialides ampulliform, (5.6-) 6.1 ± 0.4 (-6.8) µm (n = 10) (Fig. 1). Conidia globose (2.8-) 3.5 ± 0.3 (-4.0) x (2.7-) 3.4 ± 0.3 (-3.9) µm, Q = 1.02 ± 0.04, V = 21.0 ± 4.6 μm^3, n = 55, smooth to finely roughened (Fig. 2), green. Hülle cells very abundant, ovoid or elliptical, (13.3-) 19.4 ± 2.5 (-22.6) x (12.4-) 16.8 ± 1.3 (-17.00) µm, Q = 1.1 ± 0.1, n = 15, the wall (2.00-) 2.8 ± 0.4 (-3.6) µm (n = 15) thick, forming compact pale yellow masses of indefinite size, or surrounding the cleistothecia.

Cleistothecia submerged within a mycelial matrix, crowded at the colony centre, scattered to crowded in Hülle cell crusts, colour from dark blackish violet to black, globose or nearly so, 150-230 µm diameter (186 ± 17 µm, n = 30), surrounded by a loose envelope of Hülle cells and interwoven hyphae. Asci eight-spored, ovoid to globose, (9.2-) 10.7 ± 0.8 (-12.0) x (5.9-) 8.2 ± 1.3 (-10.0) µm, Q = (1.1-) 1.3 ± 0.2 (-1.7), n = 11. Ascospores deep violet-blue, deep blue, king's blue to greyish violet (M 19:B-D 7-8), lenticular, with two equatorial crests about 0.5 µm broad and 0.5 µm distant from each other, convex surfaces smooth, (3.6-) 4.2 ± 0.3 (-4.8) x (3.1-) 3.8 ± 0.2 (-4.3) x (2.8-) 3.2 ± 0.2 (-3.5) µm (n = 30); $Q_{l/w}$ = (1.0) 1.1 ± 0.1 (-1.3) (Fig. 3-4).

Colonies growing more rapidly at 37°C, attaining a diameter of more than 9 cm in 7 days. Cleistothecia formed abundantly within 7 days. No growth at 4°C.

Colonies on malt extract agar (MEA 2%, Samson et al., 1995) reaching a diameter of 3.4-4.5 cm at 25°C in 7 days, plane, centre of the colonies granular, pale yellow to light yellow (M 4: A 3-4) with a grey (M 4: B7), floccose, deep margin. Colony centre consisting of a dense layer of sclerotia with a minimum of aerial hyphae, but sometimes also without sclerotia, conidial structures scattered throughout the colony. Reverse leather to cognac to orange yellow (M 6: E 6-8), at the margin of the culture orange yellow (M 4: B7); odour negligible. Cleistothecia formed abundantly within 7 days.

Colonies on Czapek-Yeast Extract 20% Sucrose Agar (CY20S) (Klich & Pitt, 1988) thin, plane, attaining a diameter of 6-7 cm at 25°C in 7 days, velutinous, characterised by



Figs. 1-4. SEM-micrograph of the *Emericella nidulans* isolate with blue-violet ascospores. Fig. 1. Conidiophor, scale bar = 10 µm. Fig. 2. Conidia, scale bar = 1 µm. Fig. 3. Ascospores, scale bar = 2 µm. Fig. 4. Ascospore and conidium, scale bar = 1 µm.

a dense stand of conidiophores bearing columnar heads of deep green to dark green colour (M 28: E-F 7-8), few sclerotia produced. Reverse brownish-red (M 9: C-D 7-8). Odour negligible. No cleistothecia observed within 20 days.

Homothallic; ascigerous stage obtained from single conidia.

Substrate utilisation

Comparative physiological tests concerning the substrate utilisation and induction of sporulation were carried out using Biolog Microplates: our blue spored as well as an isolate of typical *Emericella nidulans* (FGSC A4 Glasgow wild type (eA+) = M139) were inoculated on MEA agar plates. From well grown cultures spore suspensions were made in sterile distilled water. The suspensions were inoculated at 100 µl per well on GN, GP and YT MicroPlates. After inoculation, the MicroPlates were incubated at 20°C for 10 days. After sufficient incubation, 'positive' reactions are determined as increased cloudiness or turbidity. In most of the positive wells, the fungi sporulate. The results were read using the Biolog MicroStation Reader and controlled by eye.

The substrate utilisation tests showed no significant differences between typical *E. nidulans* and the blue spored isolate. Both demonstrated good mycelial growth and/or sporulation on 75 substrates or substrate combinations: 2-keto-D-gluconic acid, adonitol, alpha-amino butyric acid, alpha-D-glucose, alpha-D-lactose, alpha-D-lactose+D-xylose, alpha-keto butyric acid, amygdalin, arbutin, beta-cyclodextrin, beta-methyl-D-glucoside, cellobiose, cis-aconitic acid, citric acid, D-arabitol, D-fructose, D-galactose, D-galactose+D-xylose, D-galacturonic acid, D-gluconic acid, D-glucuronic acid+ D-xylose, D-malic acid, D-mannitol, D-mannose, D-melezitose, D-melibiose, D-raffinose, D-rhamnose, D-ribose, D-saccharic acid, D-sorbitol, D-trehalose, D-xylose, dextrin, dextrin+D-xylose, fructose-6-phosphate, fumaric acid, gamma-amino butyric acid, gentiobiose, glucose-1-phosphate, glycogen, i-erythritol, L-alanine, L-alanylglycine, L-arabinose, L-asparagine, L-aspartic acid, L-glutamic acid, L-lactic acid, L-malic acid, L-phenylalanine, L-proline, L-pyroglutamic acid, L-rhamnose, L-serine, L-sorbose, L-threonine, m-inositol, m-inositol+D-xylose, maltose, maltotriose, methyl succinate+ D-xylose, mono-methyl succinate, N-acetyl-D-glucosamine, N-acetyl-L-glutamic acid+D-xylose, p-hydroxy-phenylacetic acid, palatinose, pyruvic acid, salicin, sebacic acid, stachyose, sucrose, turanose, tween 40, tween 80.

Moderate to variable growth was obtained on 68 substrates: 1,2-propanendiol+D-xylose, 2'-deoxyadenosine, 2,3-butanediol, 2-amino ethanol, 3-methyl-glucose, acetic acid, acetoin+D-xylose , alaninamide, alpha-cyclodextrin, alpha-hydroxybutyric acid, alpha-keto glutaric acid, alpha-keto valeric acid, alpha-methyl-D-galactoside, alpha-methyl-D-glucoside, alpha-methyl-D-mannoside, beta-hydroxybutyric acid, beta-methyl-D-galactoside, bromo succinic acid, D,L-alpha-glycerol phosphate, D,L-carnitine, D,L-lactic acid, D-alanine, D-arabinitol , D-arabinose , D-galactonic acid lactone, D-glucosamine , D-glucosaminic acid, D-glucuronic acid, D-lactic acid methyl ester, D-melibiose+D-xylose , D-psicose, formic acid, gamma-hydroxybutyric acid, glucose-6-phosphate, glycerol, glycyl-L-glutamic acid, hydroxy L-proline, inosine, inulin, itaconic acid, L-aspartic acid, L-erythritol, L-fucose, L-leucine, L-malic acid, L-ornithine, lactamide, lactulose, malonic acid, mannan, methyl pyruvate, methyl succinate, N-acetyl-D-mannosamine, N-acetyl-L-glutamic acid, phenyl ethylamine, propionic acid, putrescine, quinic acid, quinic acid+D-xylose, sedoheptulosan, succinamic acid, succinic acid, thymidine, uridine, uridine-5'-monophosphate, xylitol.

DISCUSSION

The character distinguishing this *Emericella nidulans* isolate from other taxa of the *Nidulantes* is the constantly blue-violet colour of the ascospores. The colour of the ascospores remained constantly blue-violet after many passages, as well as in one and the same 24-month-old culture. Moreover, the blue-violet ascospores did not change their colour into reddish or purplish when microscopically observed in acid (H_2SO_4 60%) or alkaline solutions (NH_3 25%, KOH 30%), but remained blue-violet. In contrast, the ascospore pigment in most species of the *Nidulantes* changes from purple-red to orange-red with a change from an alkaline to an acid reaction (Raper & Fennell, 1965).

The taxa of the *Emericella nidulans* group (*Emericella nidulans, E. acristata* (Fennell & Raper) Horie, *Emericella nidulans* var. *lata* Thom & Raper, *E. dentata* (Sandhu & Sandhu) Horie, *E. echinulata* (Fennell & Raper) Horie) differ primarily in ascospore morphology. Many of these originally on variety level described taxa have been recombined on species level by Horie (1980) on the basis of ascospore ornamentation. However, taxa with minor differences in ascospore morphology, e.g. *Emericella nidulans* var. *lata* having ascospores with well-developed 1.5-1.8 µm wide crests, were not recombined on species level, but up to now conserved their variety rank.
The *E. nidulans* isolate in question resembles *E. nidulans* in ascospore morphology, both having smooth valves and the crests 0.5-1.3 µm wide. SEM micrographs showed no significant differences in ascospore morphology (Fig. 3-4). Following the taxonomical concept of Horie (1980), the character of constantly blue ascospores alone can not be regarded as solid basis for a taxonomic distinction on species level: there are no significant morphological differences except the ascospore pigmentation, nor could significantly differing substrate utilisation patterns be observed. The question, whether or not the character 'ascospore pigmentation' is relevant enough for a taxonomical distinction on variety level, can not be solved on the basis of the current body of evidence (e.g. with only one isolate).

Further *Emericella* taxa with blue-violet ascospores are *E. spectabilis* M. Christensen & Raper, *E. parvathecia* (Raper & Fennell) Malloch & Cain, *E. similis* Horie, Udagawa, Abdullas & Al-Bader and *E. violacea* (Fennell & Raper) Malloch & Cain. They all differ significantly from our blue-violet-spored *Emericella* isolate: *E. spectabilis* has bigger dark violet ascomata with a diameter of 240-280 µm covered by Hülle cells, also the asci (10-14 x 10-12 µm), and the conidiophores (190-370 µm) are bigger. The ascospores are violaceous (Christensen & Raper, 1978) or orange-red to vinaceous (Samson, 1979) with two 1.0-1.2 µm wide equatorial crests. Following Samson (1979), this species resembles much *E. nidulans* except for the conidiophore size and colour and arrangement of the Hülle cells. *E. parvathecia* (Raper & Fennell) Malloch & Cain is characterised by violet-purple ascospores, but much smaller cleistothecia (30-50 µm diam.) developing very late, as well as other conidiophores and other cultural characteristics. *E. similis* can easily be distinguished by the lack of anamorph as well as the ascospore surface characterised by thick, anastomosing ribs forming an irregular reticulum (Horie et al., 1990). *E. violacea* has distinctly violet ascospores with 2 narrow crests relatively close together, the convex walls are usually echinulate, but also smooth in some isolates (Christensen & Raper, 1978). Furthermore, this species can easily be separated by the big ascospores (5.5-6.5 x 4-5 µm), the lacking or few, mostly uniseriate conidial structures, as well as the colony characters.

A key to the 5 discussed taxa of *Emericella* with blue ascospores is appended:

Key for *Emericella* species with blue-violet ascospores:

1. Cleistothecia large, 240-280 µm diam.
  Ascospore crests 1-1.2 µm wide, surfaces smooth *E. spectabilis*
1.* Cleistothecia very small, 30-50 µm diam.
  Ascospores 3.2-3.8 x 2.5-2.8 µm, crests 0.5 µm wide, surfaces smooth *E. parvathecia*
1.** Cleistothecia 100-230 µm
  2. Conidiophores numerous, ascospores valves smooth,
    ascospores 4.0-4.5 x 3.0-3.4 µm, crests 0.5 µm *E. nidulans*
  2.* Conidiophores scattered, ascospores valves mostly echinulate,
    ascospores 5.5-6.5 x 4.0-5.0 µm with very low crests *E. violacea*
  2.** Anamorph lacking
    Convex ascospores surface with thick, anastomosing ribs forming an irregular reticulum, ascospores 4.0-5.0 (5.5) x 3.0-4.0 µm *E. similis*

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LEPIOTACEAE OF FLORIDA, II. LEPIOTA S. STR., SECTION FUSCOVINACEAE

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ABSTRACT

Section *Fuscovinaceae* Bon & Candusso contains species of *Lepiota* sensu stricto with ellipsoidal to ovoid basidiospores; a pileipellis composed of filamentous hyphae, often with long, thin pilocystidia, and sometimes with an understory of much shorter, clavate elements; and clamp connections absent. Two species occurring in Florida are here recognized in section *Fuscovinaceae*: *Lepiota fuscovinacea* J.E. Lange & F.H. Moller, and *Lepiota anomala* Murrill. This is the first account of the micromorphology of *L. anomala*, and the first sectional classification of this species. *L. fuscovinacea* is reported from Florida for the first time.

INTRODUCTION

Following the recognition of segregate genera within the family Lepiotaceae Roze, the genus *Lepiota* (Pers.) Gray s. str. has received various sectional treatments. Different authorities have grappled with the problem of how to arrange species with ovoid to ellipsoidal basidiospores and a cuticle consisting of filamentous hyphae, sometimes with long, thin pilocystidia. Many authors have observed a distinction among such species, between those that have clamp connections and those that do not. The species with clamp connections have been recognized in section *Ovisporae* (J.E. Lange) Kuhner by a majority of workers, including Locquin (1945), Kuhner and Romagnesi (1978), Bon (1981), Singer (1986), Candusso and Lanzoni (1990), Franco (1994), and Akers (1997). There have been some differences, however, regarding the taxonomic disposition of the species which lack clamp connections.

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Locquin (1945) noted the absence of clamp connections in *Lepiota fuscovinacea* J.E. Lange & F.H. Moller, and proposed the new section *Anomalae* to accomodate this species as well as *Lepiota pallida* Locquin. Section *Anomalae* has since been recognized by many subsequent workers, such as Bon (1981), Singer (1986), Franco (1994), and Akers (1997). However, *Lepiota pallida* was removed from *Anomalae* by Bon (1981) and Singer (1986), because it not only possesses clamp connections, but also because it has fusiform rather than ovoid to ellipsoidal basidiospores. These authors thus modified Locquin's concept of section *Anomalae*, and recognized a number of other species there as well. Other authors, however, have not recognized this section. Kuhner and Romagnesi (1978) disregarded the presence or absence of clamp connections at the sectional level, and placed *L. fuscovinacea* and related species in *Ovisporae*. Bon and Candusso (in Candusso and Lanzoni 1990) rejected section *Anomalae* as an invalid taxon, due to lack of a published Latin diagnosis for it. They credited the presence or absence of clamp connections as an important taxonomic character, however, and proposed the new section *Fuscovinaceae* Bon & Candusso to accomodate *L. fuscovinacea* and its relatives. These authors identified section *Anomalae* as a synonym of *Fuscovinaceae* in part (*pro parte*), because they, like Singer (1986) and Bon (1981), disagreed with Locquin's placement of *L. pallida* there.

In connection with this synonymy, Bon and Candusso (in Candusso and Lanzoni 1990) identified *Anomalae* as a *nomen confusum*. Technically, this is an error because the taxonomic designation of *nomen confusum*, which refers to Article 70 of the International Code of Botanical Nomenclature, was eliminated in 1975 (Stafleu et al. 1978). This is a minor point, however, which does not alter the fact that *Anomalae* is an invalid name due to the lack of a Latin diagnosis. Section *Fuscovinaceae* is valid as published under the rules of nomenclature, and adequately resolves the taxonomic dilemma connected with section *Anomalae*. Therefore, we are following the arrangement presented in Candusso and Lanzoni (1990), and recognizing *Lepiota fuscovinacea* and allied taxa in section *Fuscovinaceae*.

There are two species which occur in Florida that belong in section *Fuscovinaceae*: *Lepiota fuscovinacea* J.E. Lange & F. H. Moller, and *Lepiota anomala* Murrill. This article presents descriptions and illustrations of the morphology of these species as they occur there. This is the first account of the micromorphology of *L. anomala*, based upon study of the type, as well as the first sectional classification of this species. It is also the first report of *L. fuscovinacea* from Florida.

MATERIALS AND METHODS

The holotype of *Lepiota anomala* was borrowed from the FLAS herbarium (University of Florida). *Lepiota fuscovinacea* was determined from a single collection, obtained in the field. Herbarium abbreviations used follow Holmgren et al. (1990).

Small tissue samples were excised, wetted in 95% ethanol, then revived in water. Sections were cut by hand and mounted in 3% KOH and Melzer's reagent, following standard mycological methods (Smith 1949; Largent et al. 1977; Singer 1986). Basidiospores were mounted in cresyl blue as well. Optical microscopy studies were carried out at 400 and 1000 X, and line drawings of micromorphological features were made using a Leitz drawing tube. Sketches of basidiomata were executed freehand. Size ranges of basidiospores, basidia, cheilocystidia and pileipellis were based on ocular micrometer measurements of at least 12-15 elements of each type from each collection studied. Wall thickness of the basidiospores is illustrated in only one example for each species depicted. Colors noted in descriptions are personal interpretations, followed by references in parentheses to approximately equivalent centroid colors of the ISCC-NBS illustrated color name charts (Kelly and Judd 1965), in which the number cited is that of the color chip in the chart, and the letter sequence denotes the name of the color in abbreviated form.

RESULTS

Lepiota fuscovinacea J.E. Lange & F.H. Moller, Flora
Agaricina Danica 5: App. 5 (1940) Fig. 1

Macromorphological features: Pileus up to 7 cm broad, convex to subexpanded, umbo broad and slight, cuticle densely fibrillose to felt-like, deep vinaceous purple, initially copious but eroding away or thinning with expansion, and disrupting into fibrillose squamules, concentrically arranged, the context whitish beneath; lamellae broad, close, cream, drying brown. Stipe up to 6.5 cm long, equal, diameter up to 5 mm, white above the annulus, cream with vinaceous purple felt-like cuticular remnants below, forming bracelet-like scales, bruising reaction with rough handling orange then reddish, finally brown; partial veil cortinate, annulus soft, flexuous-contracted, persistent, cream above, with vinaceous purple cuticular fibrils below.

Micromorphological features: Basidiospores 3.5-4.4 (-5.3) x 2.2-2.6 μm (Q = 1.81); ellipsoidal to subovoid, smooth, hilar appendix

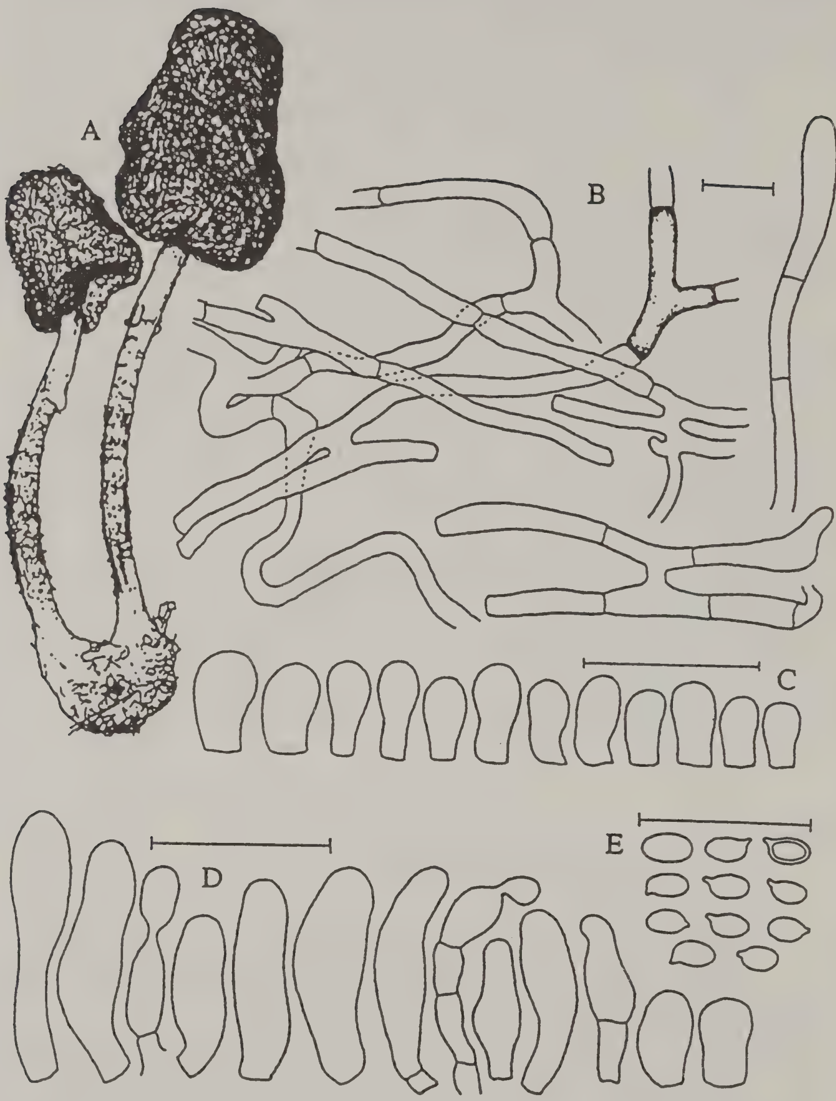

Fig. 1. *Lepiota fuscovinacea* J.E. Lange & F.H. Moller. All illustrations BPA 91-05-05-001. A. Dried basidiomata, actual size. B. Pileipellis. C. Basidioles. D. Cheilocystidia. E. Basidiospores. Scale bars = 20 µm.

prominent, germ pore none; hyaline in KOH, weakly dextrinoid, not metachromatic. Basidia not distinguished; basidioles 14-22 x 6-8.8 μm; hyaline in KOH. Cheilocystidia 10.6-30.8 x 4.4-7.5 μm, abundant, clavate to subcylindric to slightly ventricose, occasionally strangulated, often damaged, occasionally with pedicel, walls smooth, thin-sturdy; hyaline in KOH. Pleurocystidia lacking. Pileipellis of long, thin hyphae, straight to flexuous, filiform, septate to disarticulating, branching and possibly anastomosing, variably encrusted, subterminal elements 28-87 x 5-6 μm, terminal elements slightly differentiated, slightly clavate, up to 46 x 9 μm, apices rounded, vacuolar pigment golden brown in KOH. Trama below pileipellis bearing oleiferous hyphae, loosely meandering, with refractive content, amber in KOH. Clamp connections absent.

Collection studied: USA, FLORIDA. Alachua County, Gainesville, rich shaded soil near steep bank of drainage ditch on University of Florida campus, 5 May 1991, *B. Akers*, coll. and det., BPA 91-05-05-001, SIU.

Comments. The collection studied here is the only one of this species known from Florida. Originally described from Europe, this is apparently a widely distributed taxon, also previously reported from Asia and the United States (Guzman and Guzman-Davalos 1992). It seems to be rather infrequently encountered in the United States, although one of the present authors (WJS) has collected it in the Lake Itasca region of Minnesota (unpublished notes). This species has previously been described and illustrated by various authors (e.g., Locquin 1944; Reid 1967). A color photograph of it is featured in Breitenbach and Kranzlin (1995).

Lepiota anomala Murrill, Lloydia 9: 317 (1946) Fig. 2

Macromorphological features: Pileus circa 1 cm broad, conical to subexpanded, umbonate, surface dry, isabelline, darker isabelline on the umbo, minutely squamulose, margin subentire, becoming broadly striate; context thin, white, unchanging, odorless, mild; lamellae broad, ventricose, white, close, entire, unchanging; stipe equal, circa 2-3 cm long, 0.1 cm wide, pallid, fibrillose; veil white, scant, evanescent, leaving no annulus.

Micromorphological features: Basidiospores 5.1-7.9 x 3.2-4.9 μm (Q = 1.61); ellipsoidal to ovoid, smooth, hilar appendix fairly prominent, wall 0.5 μm thick, germ pore none; hyaline in KOH, moderately dextrinoid, not metachromatic although a few may display faintly. Basidia 16-25 x 7-8.5 μm; clavate, tetrasterigmate; basidioles 14-22 x 6-8.8 μm, basidia and basidoles hyaline in KOH. Cheilocystidia 19-34 x 8.5-11 μm, clavate to ventricose, occasionally with short pedicel; walls smooth, thin and sturdy; hyaline in KOH,

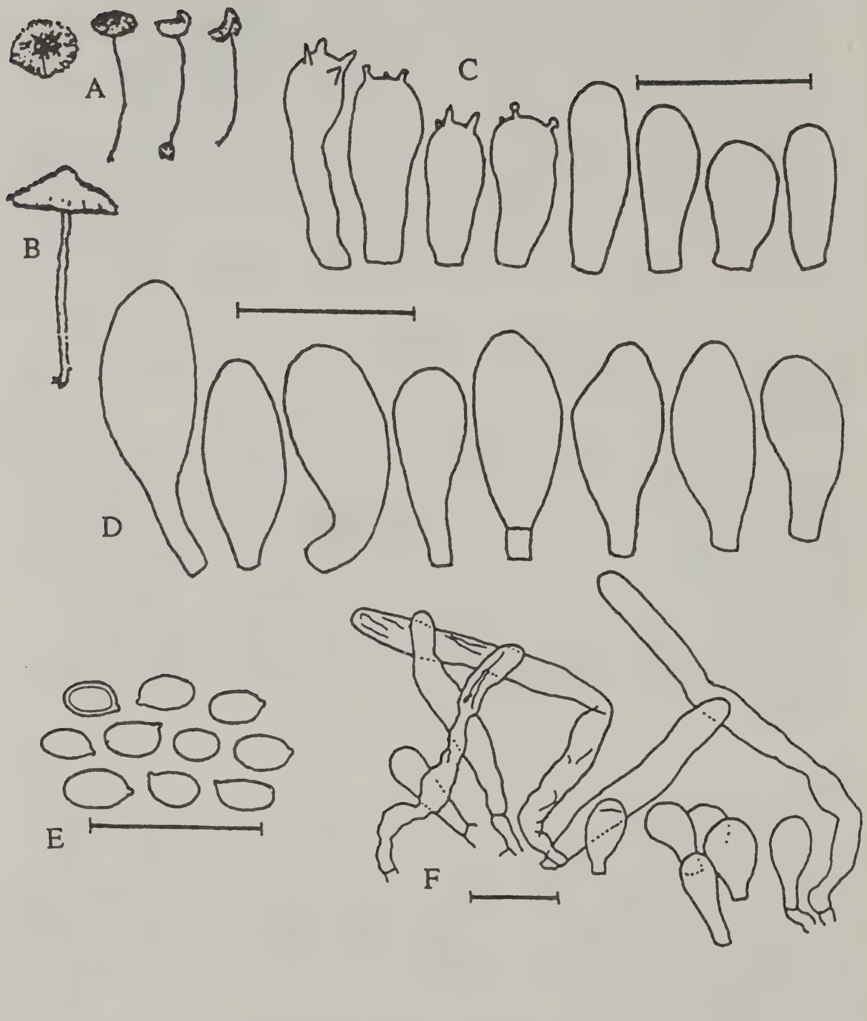

Fig. 2. *Lepiota anomala* Murrill. All illustrations FLAS F20099 (holotype). A. Dried basidiomata, actual size. B. Habit sketch by Murrill, from unpublished notes accompanying holotype. C. Basidia and basidioles. D. Cheilocystidia. E. Basidiospores. F. Pileipellis. Scale bars = 20 µm.

abundant. Pleurocystidia lacking. Pileipellis a turf of long, thin pilocystidia 59-100 x 7.5-8.5 μm, more or less erect, subcylindric with broadly rounded apices, sometimes slightly deflated and partly collapsing or falling over; sub-layer with elements 17-24.5 x 7.8-12.5 μm, clavate, reviving poorly; pilocystidia and clavate elements of sub-layer pale beige-yellow in KOH, yellow in Melzer's reagent. Clamp connections absent.

Collection studied: USA, FLORIDA. Alachua County, Gainesville, open grassy lawn, 23 Aug. 1944, *W. A. Murrill*, coll. and det., FLAS F20099 (holotype).

Comments. This is a little known species, represented only by the type collection. The data presented above on its macromorphology were taken from the original description, since no fresh collections were available for study. Murrill (1946) characterized the lamellae as close, although inspection of the exsiccatum indicated the lamellae are somewhat more distant than is typical of many other small *Lepiota* species. Murrill also indicated that the partial veil can leave delicate remnants spread along the stipe. So far, this species is known only from Florida. There seems to be no direct connection between the species epithet *anomala* and Locquin's section *Anomalae*, which was originally published in the year before Murrill described this species. The fact that this species should belong in the section bearing the same name, as a synonym, thus presents an ironic coincidence.

DISCUSSION

The species discussed here are not very well known from the United States. Considering that species in section *Ovisporae* often contain amatoxins, the close relationship between that section and *Fuscovinaceae* suggests that species in both sections should be avoided by those who would collect wild mushrooms for culinary purposes. Although some of the species in the lepiotaceous genera *Leucoagaricus* and *Macrolepiota* are well regarded as choice edible fungi, there are no species in *Lepiota* s. str. that enjoy a favorable reputation as such, nor have any local traditions of gathering them for that purpose been reported. *Lepiota fuscovinacea* has been considered as a poisonous species (Ammirati et al. 1985), but there seems to be little information available about the toxic syndrome attributed to it. A general avoidance of species in *Lepiota* s. str. seems advisable in this regard.

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INCOMPATIBILITY BETWEEN LAETIPORUS CINCINNATUS AND L. SULPHUREUS IN CULTURE

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ABSTRACT

Pairings were conducted between single basidiospore isolates obtained from 9 collections of *Laetiporus* belonging to three *Laetiporus* restriction groups (LRG). Seventy one of 81 pairings between LRGs I and VII, both of which have yellow pores and fruit on trunks or stumps of hardwoods, resulted in a change in culture morphology to a denser, more pigmented mycelium than that present in the single spore cultures. Allozyme analysis of 35 of these pairings indicated that nuclear exchange had occurred. All of the 162 pairings between LRGs I or VII and LRG IV, which has white pores and fruits on the ground near hardwoods, resulted in the formation of a dark pigmented line between the single spore isolates. No nuclear migration was indicated in any of these pairings based on allozyme analysis. These results confirm previous reports that suggest LRGs I and VII are probably the same species, *L. sulphureus*. LRG IV undoubtedly does not belong to this species but represents *L. cincinnatus*. Preliminary results of pairings between sibling single basidiospores using the culture morphology criteria suggest that both of the species have bipolar multi-allelic mating systems.

Keywords: allozyme, bipolar, isozyme, mating systems, Polyporaceae

INTRODUCTION

Laetiporus sulphureus (Fr.) Murr. in North America has referred to several forms that differ in their ecology and/or pore color. Restriction fragment length polymorphism (RFLP) analysis of the polymerase chain reaction amplified ITS region of the nuclear ribosomal DNA also shows consistent differences between these forms (Banik et al., 1996, 1998). Using the RFLP data, Banik et al. (1998) identified seven restriction groups within *L. sulphureus,* which were termed *Laetiporus* restriction groups (LRG) I-VII. The RFLPs of each of the LRGs were distinct except LRG VII appeared to be a combination of the RFLPs observed in LRGs I and VI. The restriction patterns obtained for LRG VII appeared such that they could have resulted from matings between individuals from LRG I and LRG VI. Basidiomes of all three of these groups possess yellow pore layers and fruit on stumps and trunks of hardwoods. These characteristics are generally attributable *L. sulphureus* sensu stricto.

LRG IV basidiomes possess white pore layers and occur on the ground near the base of trees, characters that coincide with the taxon described as *Polyporus cincinnatus* Morgan (Morgan, 1885). Banik et al. (1998) proposed the new combination *L. cincinnatus* (Morgan) Burdsall, Banik &Volk. In their view, the lack of combined RFLP patterns indicative of genetic exchange between LRG IV and the other LRGs, along with the distinct differences in gross morphology and ecology, substantiated the assertion that *L. cincinnatus* and *L. sulphureus* were reproductively isolated and therefore different species.

In this study, the proposed reproductive isolation between LRGs I and VII (putatively *L. sulphureus*) and LRG IV (*L. cincinnatus*) is examined using pairings between single spore isolates in culture. Because *Laetiporus* lacks clamp connections allozyme markers will be used as an independent test of nuclear migration and sexual compatibility. The compatibility of LRGs I and VII is also tested.

MATERIALS AND METHODS

Isolate collection and allozyme typing

Single spore and tissue isolates were obtained from three basidiomes each of LRGs I, IV and VII (Table 1). Basidiospores were routinely obtained by suspending sections of the pore surface over oak wood extract medium (OWEM) until basidiospores were deposited on the medium surface. OWEM was prepared by boiling 50 gr red oak sawdust in 1 L water for 1 hour and straining through two layers of cheesecloth to obtain oak wood extract. After autoclaving, 500 mL of the extract was combined with 500 mL water containing 20 gr agar. After 2-4 days incubation at room temperature, germinated single basidiospores were transferred, along with a small piece of underlying medium, to 15 x 45 mm vials containing 2 mL of OWEM. After several months the resulting single spore isolates were transferred to 16 x 125 mm culture tubes containing 1.5 % malt extract and 2 % agar (MEA) for long term storage. Tissue isolates were obtained by excising small pieces of basidiome trama onto MEA.

Tissue isolates from each of the collections were subjected to allozyme analysis for the dimeric enzyme glucose 6-phosphate (GPI) in order to obtain markers for use as aids in interpreting pairing results. A small amount of mycelium was scraped from cultures growing on medium consisting of 4 % potato dextrose agar (Difco), 0.5% agar and 0.25 % gallic acid (PDAgal) and ground in a ground glass tissue homogenizer with the addition of 100 µL extraction buffer (0.05 M Tris-HCl, pH 7.1). The resulting homogenate was centrifuged at 13800 relative centrifugal force for three minutes in a Eppendorf microcentrifuge, and the supernatant was used for allozyme analysis using cellulose acetate electrophoresis. The procedure followed the instructions provided by the manufacture of the celluose acetate gel plate electrophoresis apparatus (Helena Laboratories, Beaumont, TX) (Hebert and Beaton, 1993). The running buffer was Tris-glycine (0.25 M Trizma-base, 1.9 M glycine, pH 8.5) and gels were run for 30 min at 200 volts. Gels were stained for GPI activity using the technique of Richardson et al. (1986), modified by the addition of 2 mL of 1.5 % agar and the use of 0.1 M Tris, pH 7.4 as the stain buffer. Gels were incubated for 10 - 60 min in the dark at room temperature before being photographed and scored for allozyme activity.

Table 1. Field data for nine collections of *Laetiporus* from three *Laetiporus* restriction groups (LRG), from which basidiospores were obtained for incompatibility studies.

LRG	collection number	date collected	host	pore color	fruiting position	location
I	GR-12	10/11/95	hardwood	yellow	log	Grant Co, WI
	NJ-2	10/8/95	*Quercus* sp	yellow	trunk	Morris Co, NJ
	TJV 95-84	10/8/95	*Prunus serotina*	yellow	trunk	Dane Co, WI
IV	DA-37	10/15/97	*Q. velutina*	white	soil	Dane Co, WI
	WS-1	9/30/95	-	white	soil	Washington Co, WI
	TJV 95-49	9/3/95	*Acer saccharum*	white	soil	Sauk Co, WI
VII	CT-1	10/5/95	*Acer* sp.	yellow	stump	Darien Co, CT
	TJV 95-53	9/8/95	*Acer* sp.	yellow	trunk	Baraga Co, MI
	TJV 95-83	10/7/95	*Quercus* sp.	yellow	log	Dane Co, WI

Pairings between and within LRGs

Three single spore isolates were selected at random from each of the three collections representing either LRG I, IV or VII, for a total of 27 single spores isolates. These isolates were paired in all possible combinations on PDAgal. Two replicates of each pairing were done, one in 150 x 25 mm and one in 90 x 15 mm plastic Petri plates. Pairings were conducted by placing one agar plug, approximately 5 mm^2 cut from cultures growing on PDAgal, of each of the two isolates to be paired in contact with each other in the center of the plate. A second plug of each isolate was placed 2 cm away from the first plug, such that the four plugs in the plate were in line, with the plugs from one isolate on one side of the plate and the plugs from the other on the opposite side. After incubating at 25 C for 7 to 10 days, the morphologies of the pairings were recorded. Two basic categories of morphologies were observed, one in which the two single spore isolates appeared to fuse and the other in which the two isolates remained distinct.

Allozyme analysis was performed on 59 unique pairings in which the two single spore isolates remained distinct. Two samples from each of these pairings, one from each side, were analyzed by removing a small amount of mycelium and treating it as described earlier. Allozyme analysis was performed on 54 pairings in which the two single spores appeared to fuse by removing a small amount of mycelium from the center fused area. In all pairings analyzed, the two single spore isolates possessed different allozymes so that the detection of heterodimeric activity could be used as an indicator for nuclear combination.

Parings within collections

To determine the mating systems present in *L. cincinnatus* and *L. sulphureus*, all sibling single spore isolates from four collections were paired in all combinations using the protocol described above. Collections WS-1, DA-37 (LRG IV), TJV 95-53(LRG VII), and GR-12 (LRG-I), possessing 18,23, 20 and 15 single spore isolates respectively, were used. The pairings were incubated and scored as described above.

RESULTS

Basidiospore collection and initial allozyme analysis

Basidiospores of all collections reliably germinated on OWEM with germination percentages ranging from 20 - 80 %. Survival of the germinated basidiospore when transferrred to OWEM was close to 100 %.

Four allozyme types, designated A - D were identified in tissue isolates from the 9 collections (Fig. 1). Each of the allozyme patterns possessed only a single band, indicating that each collection was homozygous at the GPI locus. All three LRG IV collections and one LRG VII collection were type B. One collection each of LRG I and LRG VII was type A and one of each was type C. The remaining LRG I collection was type D.

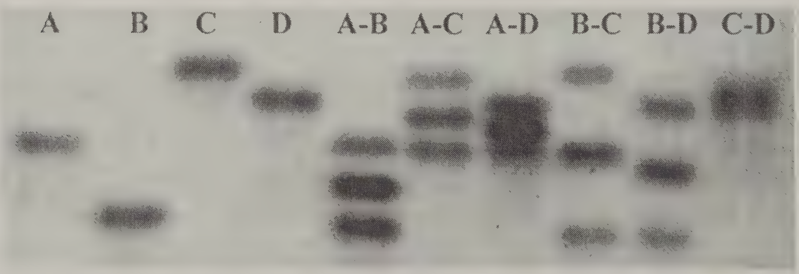

Fig. 1. Representative GPI allozyme patterns obtained from isolates of *Laetiporus* by electrophoresis on cellulose acetate gels. Lanes A-D) Homodimeric patterns from nine different tissue isolates belonging to three LRGs: type A found in LRGs I and VII, type B found in LRGs IV and VII, type C found in LRGs I and VII, type D found in LRG I. The remaining lanes are the heterodimeric patterns obtained from pairings of basidiospores of different allotypes.

Pairings within and bewteen LRGs

Within each LRG, 45 pairings were made between the 9 single spore isolates from three different collections. In LRG I, 31 of these pairings exhibited fusion (Fig. 2a) in which the center area was denser and often more pigmented than that of the mycelium resulting from the single spore isolates growing from the outer seed plugs. This included all 27 pairings between isolates from different collections. The 9 self pairings exhibited fusion without a change in morphology (Fig. 2b). In the remaining 5 pairings, the single spore isolates remained separate, although the formation of a distinct line separating them did not occur (Fig.2c). Pairings exhibiting this morphology always involved sibling basidiospores. Allozyme analysis of 7 pairings that exhibited fusion with a change to a denser, more pigmented morphology, also demonstrated heterodimeric GPI activity (Table 2, Fig. 1)

Twenty-five of the 45 pairings within LRG VII exhibited fusion with the center area becoming denser and more pigmented. Non-sibling pairings accounted for 21 of these. The 9 self pairings fused without a change in morphology as did 4 sibling pairs. Six non-sib pairs involving the same single spore isolate of collection TJV-95-53, as well as one sibling pairing with this isolate, exhibited non-fusion without a line morphology. Fourteen fusion pairings with a change in morphology were analyzed for GPI activity, and they all exhibited heterodimeric activity (Table 2, Fig 1).

Twenty-one of 36 non-sibling pairings between the three LRG IV collections expressed fusion morphology with increased density and pigmentation as did 6 sibling pairings. In the remaining 6 non-sibling pairings the two single spore isolates remained separate without the formation of a line. All of these 6 pairings involved the same single spore isolate of collection WS-1. The 9 self pairings all exhibited fusion without a change in density or pigmentation as did 2 sibling pairs. In one sibling pair the isolates remained separate without line formation (Table 2). Allozyme analysis of these pairings could not be conducted because all isolates were the same GPI allotype.

Eighty-one unique basidiospore isolate pairings were made between each two LRGs. Between LRG I and LRG VII these resulted in 71 pairings in which fusion with increased density and pigmentation occurred. Eight of the ten remaining pairings involved the same single spore isolate, and these pairings mostly remained separate without line formation as did the last two pairings. Thirty five of the pairings between LRG I and VII were subjected to allozyme analysis and all of them exhibited heterodimeric pattern types (Table 2, Fig 1). Two of these pairings exhibited the separate type morphology and samples were taken from both sides of the pairing. Heterodimeric GPI activity was detected on only one side of each of these pairings.

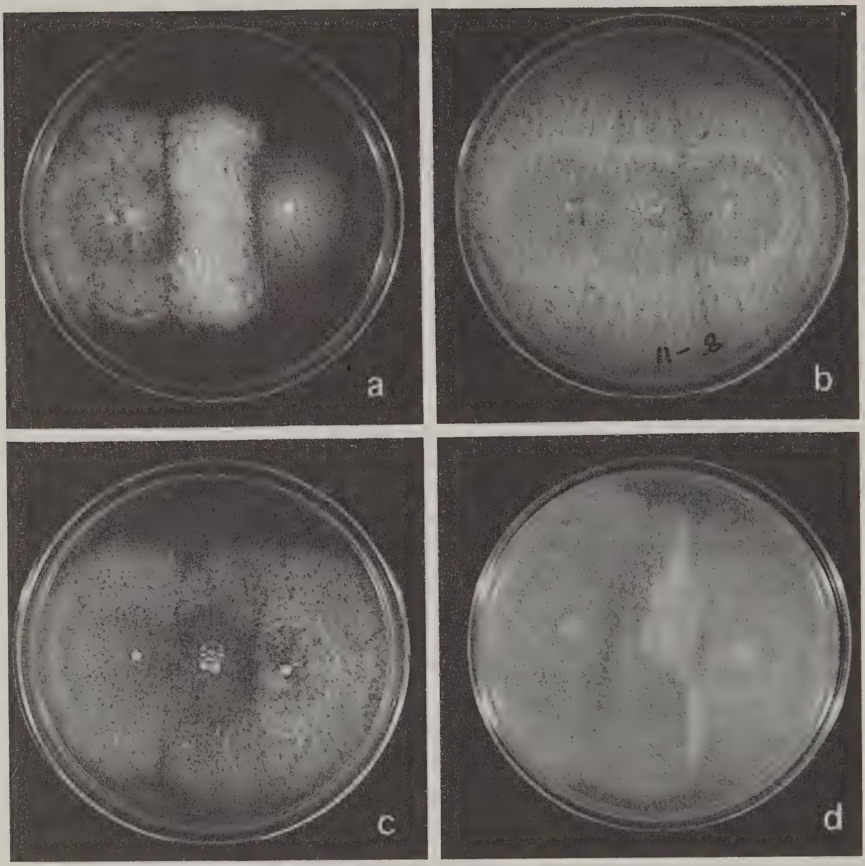

Fig. 2. Culture morphologies observed in pairings between single basidiospore isolates of *Laetiporus*: a) fusion reaction with increased density and pigmentation of mycelium, b) fusion reaction without an increase in density or pigmentation, c) separation of single spore isolates without the formation of a distinct line, d) separation of the single spore isolates with the formation of a distinct line.

Table 2. Results of pairings between basidiospore isolates of *Laetiporus* from nine collections belonging to three LRGs and frequency of recovery of heterodimeric allozyme patterns.

restriction groups paired	number of pairs	number of pairs rated with each reaction type: fusion with increased density	dense line of separation	fusion	separated without a line	number of pairings with heterodimeric GPI activity
I-I	45	31	0	9	5	7/7
I-IV	81	0	81	0	0	0/26
I-VII	81	71	0	3	7	35/35
IV-IV	45	27	0	11	7	-[a]
IV-VII	81	0	81	0	0	0/31
VII-VII	45	25	0	13	7	14/14

[a]Allozyme analysis was not done within LRG IV because all collections were the same allotype.

Pairings between either LRG I or LRG VII and LRG IV all resulted in the formation of a dense, darkly pigmented line separating the two single spore isolates (Fig. 2d). Allozyme analysis of 26 pairings between LRG IV and LRG I and 31 pairings between LRG IV and LRG VII failed to detect heterodimeric GPI activity (Table 2).

The results were the same between both replicates for 270 of 278 pairings made. All but one of the 8 pairings that did not agree were between sibling single spore isolates. These pairings were all rated as fusions without increased density or as remaining separate without line formation.

Pairings within collections

Reaction types observed in pairings of single spore isolates from the same fruiting body were the same as those observed in within-LRG pairings. None of the intracollection pairings exhibited a dense dark line as seen between LRG IV and LRG I or VII. Fusion with increased density and pigmentation (Fig. 2a) occurred in 43 %, 48 %, 51 %, and 52 % of the non-self pairings between sibling single spores of DA-37, 95-53, WS-1, and GR-12 respectively. The pattern for this morphology among the pairings fit that expected for a bipolar mating system except for 1 of the 231 DA-37 pairings, 2 of the 136 WS-1 pairings, and 2 of the 120 GR-12 pairings. In addition, one single spore isolate of collection TJV-95-53 and one of WS-1 did not express the increased density morphology in any pairings with sibling single spore isolates, and these two isolates were removed from the study. These same single spore isolates rarely or never elicited the increased density morphology in within-LRG pairings. Self pairings exhibited fusion in all instances (Fig. 2b). The non-self pairings that did not fuse with increased density of the mycelium either fused without the density change or remained separate without dark line formation (Fig. 2c). No discernible pattern accompanied the occurrence of these latter two morphologies. Because of the homozygous condition of the GPI locus for each of the basidiomes from which the basidiospores were isolated, allozyme analysis of the intracollection pairings was not possible.

DISCUSSION

The use of OWEM facilitated the collection of viable basidiospores of *Laetiporus*. Previous attempts to obtain single spore isolates using other media had been thwarted by the lack of basidiospore germination. With a reliable way to obtain basidiospores a critical assessment of the incompatibility within the genus is now possible.

The occurrence of the heterodimeric GPI activity in pairings between single spore isolates from different collections of either LRG I or VII substantiates the compatibility of these isolates. To confirm that heterodimer formation resulted from nuclear exchange and not mycelial mixing, the mycelia from ten pairs of separately grown single spore isolates were ground together and analyzed for GPI activity. The pairs of single spore isolates chosen had exhibited heterodimeric activity when they were paired during the compatibility tests. None of the ten pairs of separately grown single spore isolates exhibited heterodimeric activity when mixed during grinding. Also, samples taken from five pairings between LRG IV and LRGs I or VII exhibited the homodimeric GPI activity of each of the single spore isolates, but no heterodimeric activity was detected. In these cases, invasive growth of mycelia between the two single spore isolates occurred but nuclear exchange did not take place. Thus it appears that heterodimeric enzyme is formed only when the two isotypes are present in a biologically active thallus.

Heterodimeric GPI activity correlates consistently with the morphological change of the cultures to a denser, more pigmented state, thus substantiating that this morphology is indicative of compatibility. This type of dimorphism has been described for other fungi, such as *Armillaria mellea* (Vahl: Fr.) Kummer (Hintikka, 1973). In *Laetiporus*, the dimorphism is comparatively subtle, but by using the pairing protocol described it can be consistently and reliably used to assess compatibility.

The allozyme activity and morphological data confirm that LRG I and LRG VII are sexually compatible and represent *L. sulphureus* sensu stricto. The consistent formation of a dark line in all pairings between LRG IV and LRGs I or VII accompanied by the lack of heterodimeric GPI activity confirm that LRG IV is incompatible with *L. sulphureus*. Undoubtedly, LRG IV is a different species, and its designation as *L. cincinnatus* is appropriate. For both species, the almost complete compatibility between collections indicates the presence of multi-allelic mating system..

The pattern of compatible reactions in within-basidiome pairings fits almost exactly that expected for a bipolar mating system for both *L. sulphureus* and *L. cincinnatus*. Incompatible reactions in these crosses apparently appear either as fusions without an increase in density or a separation of the two single spores without the formation of a dark line. The two single spores isolates that did not fit the mating system pattern also did not exhibit the expected fusion with increased density morphology in within-LRG pairings. The most likely explanation for this is that these putative single spore isolates were in fact heterokaryotic. In preliminary pairing results using known heterokaryons and haploids, we have observed reactions similar to those observed with these two isolates (unpublished). Also allozyme analysis of two pairings involving one of the questionable single spore isolates suggest that nuclear migration occurred in only one direction. This might be expected if a homokaryon were paired with a heterokaryon.

The presence of a bipolar mating system is not unexpected since many other brown rot fungi have this type of mating system (Ryvarden, 1991). Confirmation of the mating system type should be possible if single spores can be obtained from basidiomes that are heterozygous at the GPI locus, making allozyme analysis of sibling pairings possible.

The conspecificity of LRG VI with *L. sulphureus* still remains to be examined. No single spores isolates of LRG VI currently exist so it was not included in this study. Also, the status of LRG II, present in the southern United States, and LRG III, which occurs on conifers, still need to be addressed. Work is currently underway to examine the compatibility among these groups and with *L. sulphureus* and *L. cincinnatus* using the techniques presented here.

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CHAENOTHECOPSIS TRISTIS, A RARE RESINICOLOUS SPECIES OF THE MYCOCALICIACEAE

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ABSTRACT

The new combination *Chaenothecopsis tristis* is suggested. The morphology of the species is described and it is compared with *Chaenothecopsis pusilla*.

INTRODUCTION

Calicium triste was described by Körber (1855) from material collected by Hampe near Blankenburg in the Harz mountains, in Germany. Körber indicated the small agglomerated apothecia and black thallus to be characteristic for the species. In a later paper (Körber 1865) he also pointed out the peculiar ecology of the species, which grew on an old *Acer* struck by the lightning. This event, in Körber's opinion had possibly caused the formation of the new species. In later treatments all references to *C. triste* were based on the type material (Massalongo 1856, Nylander 1857, Rabenhorst 1870, Sydow 1886, Saccardo 1889). That no further material was collected probably threw doubts on the species, and finally it was rejected. Nylander (1858-1860) considered it a synonym of *Phaeocalicium populneum* (Duby) A. F. W. Schmidt, and later (Zahlbruckner 1922) *Calicium triste* was considered a variety of *Chaenothecopsis pusilla* (Flörke.) A. F. W. Schmidt. However, further material identified as *Calicium triste* was collected in the Pskov province of Russia in 1982, and this makes a reevaluation possible.

MATERIAL AND METHODS

The results are based on herbarium material from LE and UPS. For scanning electron microscopy material was mounted on specimen stubs, air-dried and coated with gold.

Estimates of the apothecium height, capitulum width, stalk width, ascus length, ascus width, spore length and spore width are given in the following way: (minimum measurement recorded) a-b (maximum measurement recorded) - or for combinations of length and width: (minimum measurement recorded) a-b x c-d (maximum measurement recorded)- where a (and c) represent the arithmetic mean minus 1 standard deviation and b (and d) the arithmetic mean plus 1 standard deviation. The arithmetic mean (X) and the number of observations (n) are also supplied. The measurements have been calculated from the Pskov material.

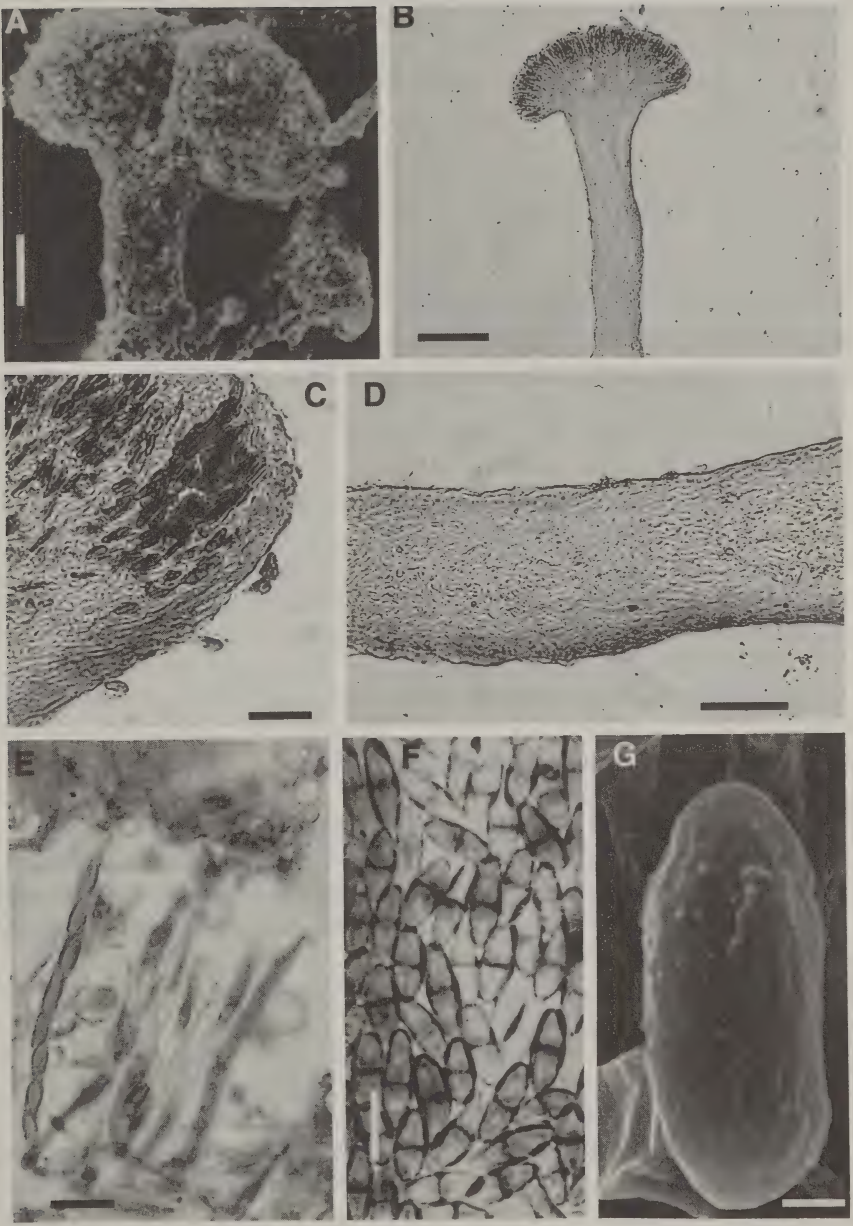
RESULTS

The Pskov material was collected on old trees destroyed by fire or lightning, just like the original material. The anatomy of the apothecia and the ascus structure indicate that the species at present is best accomodated in *Chaenothecopsis* Vain.

Chaenothecopsis tristis (Körber) Titov comb. nov. (Fig. 1).

- *Calicium triste* Körber, Syst. Lich. Germ. Breslau: 308 (1855). Type: not designated.

Fig 1. *Chaenothecopsis tristis.* Titov 727 (LE). A. Apothecium with three capitula. B. Longitudinal section of apothecium. C. Excipulum. D. Stalk. E. Semimature asci with canal in apical thickening. F - G. Mature spores. Bars: A, B = 50 µm, C = 10 µm, D = 20 µm, E = 10 µm, F = 5 µm, G = 1 µm.


A
B
C
D
E
F
G

Saprobe on exudate of bark of *Acer* and *Tilia*. Apothecia small, (0.20)0.21-0.29(0.40) mm high (X=0.25 mm, n=41), often agglomerated in groups. Stalks often branched, with several capitula. Capitulum obconical to subspherical, (0.06)0.08-0.18(0.25) mm in diam. (X=0.13 mm, n=45), black. Epithecium thin, poor developed. Epithecium, hymenium and hypothecium reddish to yellowish brown. Excipulum poorly developed, formed as a continuation of the outer layers of the stalk, brown, 5-10 µm thick. Stalk straight or curved, (0.03)0.03-0.05(0.06) mm in diam. (X=0.04 mm, n=43), dark-brown to usually pale in the lower part. The interior of the stalk pale, consisting of irregularly interwoven hyphae with strongly swollen walls and very thin lumina (c. 1 µm). The outermost part of the stalk consists of periclinally arranged, elongated cells. This surface layer is reddish to violet red and turns dark greyish violet with KOH and intensified in colour with H. Asci (30.0)46.6-52.0(60.0) x (3.0)3.3-4.0(4.0) µm (X_l=49.3 µm, n=47; X_W=3.7 µm, n=44), cylindrical or narrowly clavate. Ascus apex thickened and in young asci penetrated by a fine canal, which becomes widened in mature asci. Spores uniseriately arranged in the asci, periclinally or obliquely orientated. Spores 1-septate, (6.00)7.10-8.28(9.80) x (2.00)2.10-2.72(3.00) µm (X_l=7.68 µm, n=46; X_W=2.41 µm, n=47), ellipsoidal, medium brown, smooth.

C. tristis is only known from Germany and the European part of Russia. It was collected in open situations on exudate of bark of old trees of *Acer* and *Tilia* damaged by fire.

Material examined: Germany. Sachsen-Anhalt, Harz mountains, near Blankenburg, on bark of *Acer*, 1845 Hampe (UPS); ditto, 1851 Hampe (LE); ditto, Massalongo: Lich. Ital. exs. 353 (UPS). Russia. Pskov province, Strugi Krasnye region, Tvorozhkovo, on *Tilia cordata*, 1982 Titov 727 (LE, UPS).

DISCUSSION

Chaenothecopsis tristis is characterized by its agglomerated apothecia, the 1-septate smooth spores, apothecium anatomy, and the occurrence on exudate of deciduous trees. This species may be mistaken for *Chaenothecopsis pusilla*, which is very variable and possibly in the present circumscription includes several species (Tibell 1987). *Chaenothecopsis tristis* differs from *C. pusilla* by having larger apothecia, asci and spores, a reddish epithecium and excipulum, and a reddish KOH+ and H+ intensified reaction of the stalks. *C. tristis* occurs on exudate of bark of deciduous trees where neither lichens nor free-living colonies of algae occur, whereas *C. pusilla* primarily occurs on lignum of variety of trees and often together with calicioid lichens and free-living algae.

Tibell and Titov (1995) recently published a world-wide revision of species of Mycocaliciaceae growing on exudate of vascular plants. Eight species occurring in the Russian Far East, Caucasus, India, the Americas and New Zealand were included, but the majority of them have only rarely been collected. *C. tristis* also belongs to this ecological group of fungi, and like the other species of this group may have been overlooked by collectors.

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NEW RECORDS OF PHYLLACHORA ON POACEAE FROM ARGENTINA

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Abstract

Forty-one specimens representing fifteen species of *Phyllachora* were collected in Argentina. Seven species, *P. chusqueae, P. luteo-maculata, P. oxyspora, P. punctum, P spartinae* , *P. sylvatica* and *P. vulgata* are newly reported for Argentina and twenty-one species of grasses are new hosts for the fungus. The first collections from Patagonia are reported.

Key Words: Argentina, *Phyllachora*, Poaceae

Fungi in the genus *Phyllachora* are obligate host specific pathogens and have been reported from grasses worldwide (Parbery, 1967, 1971). Numerous species of *Phyllachora* on Poaceae have been reported from South America (Chardón, 1932; Chardón, et al. 1940; Chardón and Toro, 1930 and 1934; Da Silva and Minter, 1995; Dennis, 1970; Farr, 1973; Orejuela, 1944; Orton, 1944; Parbery, 1967 and 1971; Rada and Stevenson, 1942; Spegazzini, 1910; Stevens, 1923 and 1931; Stevenson and Cárdenas, 1949; Theissen and Sydow, 1915; Viégas, 1944 and 1961). Parbery (1967, 1971) monographed the genus on Poaceae, synonymized numerous species and compiled a list of twelve legitimate species from Argentina (Table 1). The literature indicates the distribution of *Phyllachora* on Poaceae in Argentina is poorly known and no recent work has been done. This study was initiated to investigate the present occurrence of *Phyllachora* species on grasses in Argentina and to document the current geographic distribution of these fungi.

Materials and Methods

Grasses of the pampa, forests, pastures, fields and roadside ditches were examined at frequent intervals from 13 January to 2 March 1996, along a 13,600 km. route in the following provinces: Buenos Aires, Catamarca, Córdoba, Chubut, Entre Ríos, La Pampa, Mendoza, Neuquén, Río Negro, Santa Cruz, Santa Fe, San Luis,

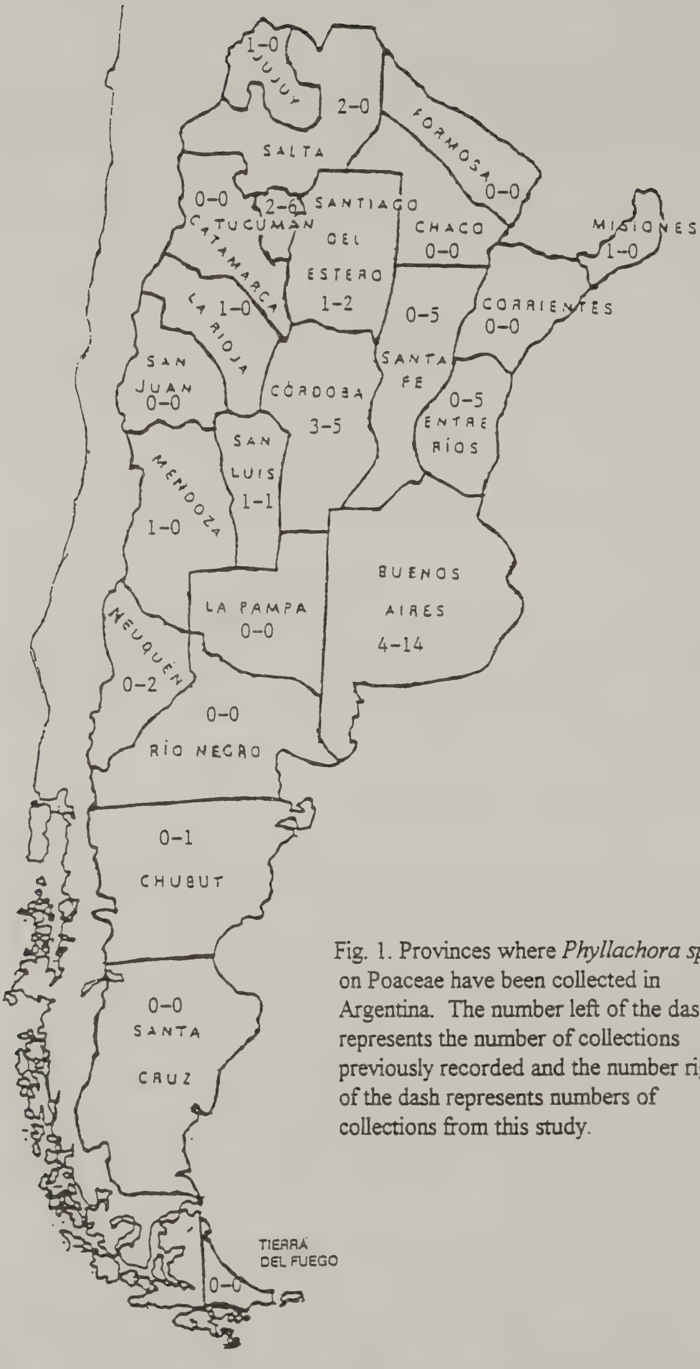
1-0
JUJUY
2-0
SALTA
FORMOSA
0-0
0-0
2-6
SANTIAGO
DEL
ESTERO
1-2
TUCUMAN
CATAMARCA
CHACO
0-0
MISIONES
1-0
LA RIOJA
1-0
0-5
CORRIENTES
0-0
SAN
JUAN
0-0
CÓRDOBA
3-5
SANTA
FE
0-5
ENTRE
RÍOS
SAN
LUIS
1-1
MENDOZA
1-0
BUENOS
AIRES
4-14
LA PAMPA
0-0
NEUQUÉN
0-2
0-0
RÍO NEGRO
0-1
CHUBUT
0-0
SANTA
CRUZ
TIERRA
DEL FUEGO
0-0

Fig. 1. Provinces where *Phyllachora spp.* on Poaceae have been collected in Argentina. The number left of the dash represents the number of collections previously recorded and the number right of the dash represents numbers of collections from this study.

Santiago del Estero and Tucumán. These provinces lie approximately between 27 and 51 degrees south latitude. Infected leaves were removed, pressed and dried in the field for later microscopic examination. Plants, including inflorescences, were pressed and dried. Taxonomic treatments by Parbery (1967, 1971) and Orton (1944) were used for identification of *Phyllachora* species. Size and shape of clypei were reported for individual clypei even though most clypei coalesced. Clypeus size was based on (5)-14-(24) observations and shape followed Hawksworth (1995). Perithecial measurements were based on (2)-9-(15) observations/specimen. Length of the sporogenous portion of the ascus and width were based on (3)-12-(24) observations/specimen. Pedicels if present were measured and lengths are listed separately. Ascospore size was based on (8)-18-(27) observations/specimen and shapes were interpreted following Parbery (1967). For most specimens spermagonia were uncommon, but were measured when present. Spermatial size was based on (7)-16-(23) observations/specimen. Grass

Table 1. Previous reports of species of *Phyllachora*, the grass host and province where collected in Argentina (Parbery, 1967 and 1971).

Phyllachora Species	Grass Species	Province
P. acutispora Speg.	*Paspalum* L.	Salta
P. antarctica Speg.	*Poa lilloi* Hackel	Tucumán
P. cynodontis (Sacc.) Niessl	*Bouteloua curtipendula* (Michx.) Torr.	unknown
	Chloris radiata (L.) Swartz	La Rioja
	Chloris Swartz	Córdoba & Tucumán
	Chloris castilloniana Lillo & Parodi	Jujuy
P. eragrostidis Chardón	*Eleusine tristachya* (Lam.) Lamarack	unknown
P. eriochloae Speg.	*Eriochloa procera*[1] (Retz.) C.E. Hubbard	Santiago del Estero
P. fuscescens Speg.	*Agrostis* L.	Buenos Aires
P. koondrookensis Parbery	*Trichloris crinata* (Lag.) Parodi	Mendoza
	Trichloris Fournier ex Bentham	Córdoba
P. minutissima (Welw. & Curr.) G. Sm.	*Echinochloa polystachya* (H.B.K.) Hitch.	unknown
P. oblongospora Parbery	*Chloris distichophylla* Lag.	Córdoba
P. sacchari Henn.	unknown	unknown
P. setariecola Speg.	*Lasiacis divaricatus* (L.) Hitchcock	unknown
P. urvilliana Speg.	*Panicum urvilleanum* Kunth	unknown

[1] = A doubtful grass host. This species is native to Fiji and not reported from Argentina.

identification followed taxonomic treatments of Burkart (1969), Cabrera (1970), Killeen (1989) and Nicora (1978). All specimens have been deposited in the Herbario, Universadad de Buenos Aires, Ciudad Universitaria, Buenos Aires (BAFC).

Exsiccatae in BAFC and Herbario, Instituto de Botánica Carlos Spegazzini, Universidad Nacional de La Plata (LPS) were also examined for specimens of *Phyllachora* on Poaceae collected from Argentina.

Results

Examination of specimens at BAFC and LPS included the same species listed by Parbery (1967, 1971) and one additional species, *P. sphaerosperma* Wint.. on *Cenchrus myosuroides* H.B.K. collected from Salta. LPS exsiccatae showed *P. cynodontis* was also collected on *Neobouteloua lophiostachya* (Griseb.) Gould (=*Bouteloua lophiostachya)* from San Luis, *P. minutissima* was also collected on *Panicum grumosum* Nees and P. *sacchari* was collected on *Sorghum vulgare* var. *saccharatum* (L.) Boerl. (= *Andropogon saccharatus)* Raspail from Misiones. The *P. eragrostidis* and *P. urvilliana* collections were from Buenos Aires. Following are listed alphabetically the species of *Phyllachora* collected in the present study, descriptions, grass hosts and collection sites.

Phyllachora acutispora Speg.

On *Paspalum dilatatum* Poiret.: Amphigenous, adaxially raised lesions on living leaves. Clypeus round to cylindrical, (.5)-1-(2) X (.25)-.4-(.75) mm, surrounded by a chlorotic to orange halo .5-1.0 mm wide. Perithecia (143)-184-(239) µm wide X (130)-170-(196) µm high. Paraphyses hyaline, filiform, 2.5 µm wide. Asci cylindrical, uniseriate to biseriate, (45)-67-(78) X (9)-10.6-(12.5) µm, eight-spored, pedicels (7.5)-14.3-(35) µm. Ascospores hyaline, one-celled, strongly ovate-acuminate, (15)-19-(23) X (5)-5.1-(6.3) µm, smooth to rugose. Spermagonia rare, 142 µm wide X 124 µm high. Spermatia hyaline, filiform, one-celled, (10)-14-(17.5) X 1.25 µm. Specimen examined: A. and M. Gabel, # 2809, Talavera Island along Rt. 12 in Río Paraná, 13-1-1996, between Provincia Entre Ríos and Buenos Aires.

On *Paspalum intermedium* Munro ex Morong et Britton: Amphigenous raised lesions on living leaves. Clypeus oval to cylindrical, (.75)-.9-(2.0) mm X (.25)-.4-(.5) mm, surrounded by a yellow halo, .25-1.0 mm. Perithecia (130)-165-(200) µm wide X (130)-163-(209) µm high. Paraphyses hyaline, filiform, 1.3-2.5 µm wide. Asci cylindrical to clavate, biseriate (62.5)-82-(115) X (10)-15-(20) µm, eight-spored, pedicels not observed. Ascospores hyaline, one-celled, elongate to acuminate, (15)-19.7-(24) X (5)-6.1-(6.3) µm, smooth. Spermagonia numerous, (44)-112-(174) µm wide X (109)-155-(196) µm high. Spermatia numerous, hyaline, filiform, one-celled, (17.5)-22-(26.3) X 1.25 µm. Specimen examined: A. and M. Gabel, # 2903, Rt. 86 northwest of Laprida, 28-2-1996, Provincia Buenos Aires.

Comments: The acuminate shape of the ascospores is distinct and agrees with the description of *P. acuminata* Starb., which is now *P. acutispora* . The fungus has been described on *Paspalum* sp. from Argentina (Parbery, 1967, 1971). This is the first report on *P. dilatatum* and *P. intermedium.*

Phyllachora antarctica Speg.

On *Poa bonariensis* (Lam.) Kunth: Amphigenous raised lesions on living leaves. Clypeus cylindrical to filiform (1)-1.4-(2.5) X (.13)-.2-(.25) mm, with little or no chlorotic halo. Perithecia (130)-206-(283) µm wide X (131)-211-(283) µm high.

Abundant, septate, filiform paraphyses, 1.3 µm wide. Asci cylindrical, uniseriate, (78)-92-(113) X (10)-13-(16) µm, eight-spored, pedicels (5)-14-(20) µm. Ascospores dilute yellow, one-celled, oval to ovoid, (12.5)-13-(15) X (7.5)-8.6-(9) µm, smooth. Spermagonia rare, 87 X 87 µm. Spermatia hyaline, one-celled, filiform, (12.5)-16-(20) X (1.3)-1.9-(2.5) µm. Specimen examined: A. and M. Gabel, # 2856, off Rt. 20 6.0 km east of El Cóndor, in Sierra de Comechingones, Provincia Córdoba, 24-1-1996.

Comments: This species has been reported on *Poa lilloi* Hackel in Argentina (Parbery, 1971). This is the first report on *P. bonariensis.*

Phyllachora chusqueae Henn. & Lindau

On *Chusquea culeou* Desvaux: Adaxial lesions on living leaves, raised on both epidermes. Clypeus round to cylindrical, (.75)-1.5-(3.5) X (.5)-.8-(1) mm, brown discoloration 2 mm beyond groups of clypei. Perithecia (217)-361-(478) µm wide X (100)-223-(304) µm high. Sparse hyaline, filiform paraphyses 2.5 µm wide. Asci clavate, some with a subacute apex, all biseriate, (58)-86-(123) X (13)-17-(19) µm, eight crowded spores, pedicels (10)-18-(35) µm. Ascospores hyaline, one-celled, fusiform to oval, (12.5)-17.5-(20) X (6.3)-7.5-(8.8) µm, smooth. Spermagonia and spermatia were not observed. Specimens examined: A. and M. Gabel, # 2880, Parque Nacional Los Alerces, 8-2-1996, Provincia Chubut; # 2871, Parque Nacional Lanín, 2-2-1996, Provincia Neuquén.

On *Chusquea lorentziana* Griseb.: Adaxial, raised lesions on living leaves. Clypeus elongate to cylindrical, (1)-1.4-(2.8) X (.75)-.8-(1) mm, with little or no halo. Perithecia (239)-287-(348) µm wide X (100)-171-(261) µm high. Abundant hyaline, filiform paraphyses, 2.5 µm wide. Asci clavate, biseriate, (83)-99-(130) X (15)-22-(25) µm, eight crowded spores, pedicels (10)-8-(37) µm. Ascospores hyaline, fusiform to oval, (20)-24-(27.5) X (7.5)-9.4-(12.5) µm, smooth. Intact spermagonia not observed. Spermatia hyaline, filiform, one-celled, (15)-19-(22.5) X 2.5 µm. Specimen examined: A. and M. Gabel, #2844, 1.3 km west of Río de la Sosas, near Tafí, 22-1-1996, Provincia Tucumán.

Comments: The clavate to subclavate ascus with a subacute apex agrees with the Spegazzini (1910) description. All asci were biseriate as described by Parbery (1967) and Spegazzini (1910). Although ascospores from *C. lorentziana* are larger than those on *C. culeou* and those reported by Parbery (1967), all other characters are idential and the fungus is considered the same species. The fungus has been described on *Chusquea* sp. from Chile (Parbery, 1967; Spegazzini, 1910). This is the first report of the fungus in Argentina and the first report on *C. culeou* and *C. lorentziana.*

Phyllachora cynodontis (Sacc.) Niessl

On *Cynodon dactylon* (L.) Persoon: Amphigenous, raised lesions on living leaves. Clypeus round to cylindrical, (.13)-.75-(1.5) X (.1)-.52-(1) mm surrounded by yellow to red halo (.25-.75 mm). Perithecia variable in size and shape, constricted by vascular bundles, (65)-121-(239) µm wide X (65)-110-(165) µm high. Uniseriate asci cylindrical, more common biseriate asci clavate, (40)-54-(63) X (5)-10-(13) µm, eight-spored, pedicels (5)-10-(13) µm. Ascospores hyaline, oval to ovoid, (7.5)-10.3-(12.5) X (5)-5.6-(7.5) µm, smooth. Spermagonia rare, 66 µm wide X 76 µm high. Spermatia hyaline, filiform, one-celled, (7.5)-10.6-(12.5) X (1.3)-1.3-(2.5) µm. Specimens examined: A. and M. Gabel, # 2911, Rt. 226 on way to Bosch, near Cinco Cerros, north of Balcarce, 29-2-1996; # 2914, Rt. 74 east of Ayachucho and 16.5 km east of intersection with Rt 2 at Las Armas, 1-3-1996; # 2915, 6.0 km east of turnoff at Rt. 56 and about 10 km south of General Lavalle, Provincia Buenos Aires.

Comments: Asci sporogenous lengths and spermatia of the current specimens are slightly shorter than those listed by Parbery (1967). The fungus has been reported on *Bouteloua* and *Chloris radiata* (L.) Sw. from Argentina, and on *Cynodon dactylon* from

Uruguay (Parbery, 1967). This is the first report on *C. dactylon* in Argentina.

Phyllachora eragrostidis Chardón

On *Eleusine indica* (L.) Gaertner: Amphigenous, raised lesions on living leaves. Clypeus round to oval, (.5)-1-(2) X (.5)-.9-(1) mm, with little or no halo. Perithecia (178)-237-(294) µm wide X (147)-177-(210) µm high. Hyaline, filiform paraphyses, 1.3 µm wide. Asci cylindrical, uniseriate, (63)-79-(88) X (7.5)-8-(10) µm, eight-spored, pedicels (5)-11-(25) µm. Ascospores ellipsoid, (7.5)-10.6-(13.8) X (5)-5.9-(7.5) µm, smooth. Spermagonia infrequent, 174 µm wide X 96 µm high. Spermatia hyaline, filiform, one-celled, slightly curved, (10)-12.6-(20) X 1.3 µm. Specimen examined: A. and M. Gabel, # 2824, Isla El Timbo between cities of Paraná and Santa Fe, 14-1-1996, Provincia Entre Ríos.

On *Eragrostis lugens* Nees: Amphigenous, raised lesions on living leaves. Clypeus round to cylindrical, (.5)-1.6-(6) X (.25)-.55-(1) mm, yellow halo extending to 1.0 mm. Perithecia (87)-143-(239) µm wide X (87)-141-(217) µm high. Hyaline, filiform paraphyses, 1.3 µm wide. Asci cylindrical, uniseriate, (57)-72-(88) X (7.5)-9.3-(12.5) µm, eight-spored, pedicels (8)-9-(13) µm. Ascospores hyaline, one-celled, oval to ellipsoid to ovoid, (10)-11.5-(13.8) X (5)-5.8-(9) µm, smooth. Spermagonia rare, (61)-77-(96) µm wide X (43)-51-(65) µm high. Spermatia infrequent, hyaline, filiform, one-celled, (10)-13.3-(15) X 1.3 µm. Specimens examined: A. and M. Gabel, # 2806, Talavera Island off Rt. 12, 13-1-1996, between Provincia Entre Ríos and Buenos Aires; # 2822, Isla El Timbo between cities of Paraná and Santa Fe, 14-1-1996, Provincia Entre Ríos; # 2853, Rt. 20 west of Córdoba, southwest of Villa Carlos Paz and 14.7 km. east of Copina turnoff, 24-1-1996, Provincia Córdoba; # 2901, Parque Provincial Ernesto Tornquist between Tornquist and Sierra de la Ventana, 27-2-1996, Provincia Buenos Aires.

Comments: The species has been reported on *Eleusine tristachya* Kunth from Argentina (Parbery, 1967). This is the first report on *Eleusine indica* and *Eragrostis lugens* in Argentina.

Phyllachora luteo-maculata (Schw.) Orton

On *Bothriochloa laguroides*(D.C.) Herter (= *Andropogon laguroides*): Amphigenous, raised lesions on living leaves. Clypeus cylindrical, (1)-1.7-(4) X (.25)-.5-(.75) mm with thin orange halo. Perithecia (131)-167-(217) µm wide X (131)-143-(174) µm high. Paraphyses filiform, hyaline, 1.25 µm wide. Asci cylindrical, uniseriate, (63)-72-(78) X (7.5)-7.8-(10) µm, eight-spored, pedicels (7.5)-7.5-(7.5) µm. Ascospores hyaline, one-celled, oval to ellipsoid, (8.8)-11.1-(12.5) X (5)-5.1-(6.3) µm, smooth. Spermagonia and spermatia were not observed. Specimen examined: A. and M. Gabel, # 2831, Rt 4, 3.0 km south of La Pelada, northwest of Santa Fe, 15-1-1996, Provincia Santa Fe.

On *Schizachyrium microstachyum* (Desv.) Roseng. et al.: Amphigenous, raised lesions on living leaves. Clypeus round to cylindrical, (1)-1.5-(2) X (.25)-.67-(1) mm, surrounded by a chlorotic halo, .5-1.0 mm. Perithecia (152)-203-(261) µm wide X (109)-146-(217) µm high. Paraphyses hyaline, filiform, 1.25 µm wide. Asci cylindrical, uniseriate, (60)-70-(80) X (7.5)-9-(11.3) µm, eight-spored, pedicels (5)-10-(12.5) µm. Ascospores hyaline, one-celled, oval to ellipsoid, (7.5)-9.5-(11.3) X (5)-5.5-(6.3) µm, smooth. Spermagonia uncommon, (65)-80-(87) µm wide X (52)-82-(131) µm high. Spermatia hyaline, one-celled, filiform and slightly curved, (14)-17-(20) X 1.25 µm. Specimen examined: A. and M. Gabel, # 2839, Rt. 307 to Tafi, 2 km east of La Ramadita, 18-1-1996, Provincia Tucumán.

Comments: Ascospore shape and size are similar, but asci of the current specimen, *Bothriochloa laguroides* are smaller than those described for *Phyllachora luteo-maculata* by Parbery (l967) and Orton (1944). Material of the current specimen may be immature. This is the first report of the fungus in Argentina and the first report on *B. laguroides* and on *S. microstachyum*.

Phyllachora minutissima (Welw. & Curr.) G. Sm.

On *Echinochloa helodes* (Hack.) Parodi: Amphigenous, raised lesions on living leaves. Clypeus round to elongate, (.25)-.75-(1) X (.25)-.7-(1) mm. Perithecia (109)-149-(218) µm wide X (109)-129-(174) µm high. Paraphyses hyaline, filiform, 2.5 µm wide. Asci cylindrical to clavate, mostly biseriate, (63)-72-(85) X (12.5)-14.2-(16) µm, eight-spored, pedicels not observed. Ascospores hyaline, one-celled, oval to ovoid, (12.5)-15.5-(17.5) X (5)-5.7-(7.5) µm, smooth. Spermagonia and spermatia not observed. Specimen examined: A. and M. Gabel, # 2833, 3.9 km east of Santiago Del Estero border with Santa Fe, between Ceres and Selva on Rt 34, 15-1-1996, Provincia Santa Fe.

On *Paspalum quadrifarium* Lamarck: Amphigenous, raised lesions on living leaves. Clypeus round, elongate to cylindrical, (0.5)-1.3-(5.0) X (.13)-.62-(1.75) mm, tan to yellow halo extending .25-1.0 mm. Perithecia (139)-240-(391) µm wide X (113)-200-(305) µm high. Paraphyses hyaline, filiform, 1.25-2.5 µm wide. Asci cylindrical, mostly uniseriate, (80)-105.4-(135) X (8.8)-11.9-(17.5) µm, eight spored, pedicels (5)-18.8-(62.5) µm. Ascospores hyaline, one-celled, oval to ellipsoid, a few constricted in the center, (13.8)-16.7-(20) X (5)-6.9-(8.8) µm, smooth. Spermagonia rarely observed, (174)-195.8-(239) µm wide X (196)-217.4-(261) µm high. Spermatia abundant, hyaline, filiform, straight to curved, (12.5)-19.8-(25) X 1.25 µm. Specimens examined: A. and M. Gabel, # 2908, 7.0 km off of Rt. 226, north Balcarce, 29-2-1996; # 2907, along Circuito Turístico, southwest of Tandil, 28-2-1996; # 2893, along A. Sauce Grande, Villa Arcadia, Rt.72, 27-2-1996; # 2894, golf course in Villa Arcadia, Rt. 72, 27-2-1996; # 2899, Rt. 76 east of Tornquist, 27-2-1996; Provincia Buenos Aires; # 2854, Rt. 20 southwest of Villa Carlos Pas, 24-1-1996, Provincia Córdoba.

Comments: Parbery (1967, 1971) listed the fungus on *Echinochloa polystachya* (H. B. K.) Hitch. and on *Panicum grumosum* Nees from Argentina, and on *Paspalum dilatatum* from Uruguay. These are the first reports on *E. helodes* and *Paspalum quadrifarium* from Argentina.

Phyllachora oxyspora Starbäck

On *Sorghastrum pellitum* (Hackel) Parodi: Lesions on adaxial surface of living leaves. Clypeus oval to cylindrical, (1)-1.2-(2) X (.25)-.45-(.5) mm, halo only between groups. Perithecia (131)-173-(204) µm wide X (117)-141-(152) µm high. Asci cylindrical to clavate, biseriate, (58)-72-(100) X (13)-15.5-(18) µm, eight crowded spores, pedicels (5)-19-(37.5) µm. Ascospores hyaline, one-celled, oval to ellipsoid to ovoid, (13.8)-15.5-(18.8) X (5)-6.9-(7.5) µm, smooth. Spermagonia rare, lobed, 109 µm wide X 131 µm high. Spermatia abundant, hyaline, filiform, slightly curved, (15)-17.4-(22.5) X 1.9 µm. Specimen examined: A. and M. Gabel, # 2909, 7.0 km from Rt. 226 north of Balcarce on road to Bosch, near Cinco Cerros, 29-2-1996, Provincia Buenos Aires.

Comments: The fungus has been reported on *Sorghastrum* and from South America (Parberry, 1967). This is the first report of the fungus in Argentina and the first report on *S. pellitum*..

Phyllachora punctum (Schw.) Orton & F. Stevens

On *Paspalum lividum* Trinius ex Schlechtendal: Amphigenous raised lesions on living leaves. Clypeus round to cylindrical, (.5)-.83-(1.5) X (.25)-.43-(1) mm, surrounded by an orange halo (.13-.5 mm). Perithecia (87)-145-(217) µm wide X (87)-123-(196) µm high. Paraphyses hyaline, filiform, 1.3-2.5 µm wide. Asci cylindrical, uniseriate, (62.5)-79-(105) X (9)-10-(12.5) µm, eight-spored, pedicels not observed. Ascospores hyaline, one-celled, oval, (11.3)-12.5-(15) X (5)-5.4-(6.3) µm, smooth. Spermagonia and spermatia not observed. Specimen examined: A. and M. Gabel, # 2832, Rt 4, 3 km south of La Pelada, 15-1-1996, Provincia Santa Fe.

On *Paspalum malacophylum* var. *linearifolium* Hackel en Fedde: Amphigenous raised lesions on living leaves. Clypeus round to cylindrical, (.75)-1.45-(2.0) X (0.5)-.85-(1.0) mm, surrounded by minimal discoloration. Perithecia (139)-203-(313) µm wide X (139)-166-(196) µm high. Paraphyses hyaline, filiform, 2.5 µm wide. Asci cylindrical, uniseriate to biseriate, (60)-76-(102) X (7.5)-9.1-(10) µm, eight-spored, pedicels (10)-14.4-(20) µm. Ascospores hyaline, one-celled, oval to ovoid, (11.3)-13-(16.3) X (5)-5.2-(6.3) µm, smooth. Spermongonia not observed. Spermatia hyaline, filiform, straight to curved, (12.5)-15-(17.5) X 1.25 µm. Specimen examined: A. and M. Gabel, # 2836, 5.0 km south Rt 9 near Termas Río Hondo, 16-1-1996, Provincia Santiago Del Estero.

On *Papalum nicorae* Parodi: Amphigenous, adaxially raised lesions on living leaves. Clypeus oval to cylindrical, (1)-1.3-(2) X (.25)-.39-(.5) mm, surrounded by an orange-tan halo, .5-1.0 mm. Perithecia (161)-221-(239) µm wide X (122)-196-(226) µm high. Paraphyses hyaline, filiform, 2.5 µm wide. Asci cylindrical, uniseriate, (63)-79-(100) X (7.5)-10-(15) µm, eight-spored, pedicels (7.5)-10-(12). Ascospores hyaline, one-celled, oval, (12.5)-13.5-(17.5) X (5)-6.4-(7.5) µm, smooth. Spermagonia rare, 174 µm wide X 148 µm high. Spermatia hyaline, filiform, one-celled, straight to slightly curved, (10)-17-(22.5) X 1.25 µm. Specimen examined: A. and M. Gabel, # 2823, Isla El Timo along Río Paraná, 14-1-1996, Provincia Entre Ríos.

On *Paspalum notatum* Fluegge: Amphigenous, raised lesions on living leaves. Clypeus round to cylindrical, (.25)-1.1-(3) X (.25)-.6-(1) mm, surrounded by purple-orange halo, .5-1 mm wide. Perithecia (130)-208-(283) µm wide X (130)-177-(261) µm high. Paraphyses hyaline, filiform, 1.25-2.5 µm wide. Asci cylindrical, uniseriate to partially biseriate, (63)-85-(125) X (7.5)-10.9-(13.8) µm, eight-spored, pedicels (5)-17-(50) µm. Ascospores hyaline, one-celled, ovoid to ellipsoid, (11.3)-13.5-(17.5) X (5)-5.6-(7.5) µm, smooth. Spermagonia not observed. Spermatia hyaline, filiform, one-celled, straight to curved, (10)-15-(18) X 1.25 µm. Specimens examined: A. and M. Gabel, # 2852, Rt. 20 southwest of Villa Carlos Paz, 24-1-1996, Provincia Córdoba; # 2840, Rt. 307 east of Tafí del Valle along Río de la Sosas, 18-1-1996; # 2846, Rt. 307 east of Tafí del Valle, west of junction with Rt. 324, 22-1-1996, Provincia Tucumán; # 2837, 5.0 km south of junction of Rt. 9 and Rt 333 near Termas Río Hondo, 16-1-1996, Provincia Santiago Del Estero.

On *Paspalum plicatulum* Michx.: Amphigenous, raised lesions on living leaves. Clypeus round to cylindrical, (.75)-1.2-(3) X (.25)-.6-(1) mm, surrounded by chlorotic halo .5-1 mm. Perithecia (109)-202-(348) µm wide X (109)-187-(304) µm high. Paraphyses hyaline, filiform, 2.5 µm wide. Asci cylindrical, uniseriate to partially biseriate, (60)-84-(120) X (7.5)-10.4-(13.8) µm, eight-spored, pedicels (7.5)-17-(30) µm. Ascospores hyaline, one-celled, oval to less commonly ovate-acuminate, (11.3)-12.9-(17.5) X (4)-5.6-(7.5) µm, smooth. Spermagonia rare, 120 µm wide X 196 µm high. Spermatia hyaline, one-celled, straight to slightly curved, (12.5)-14.4-(20) X 1.25 µm. Specimens examined: A. and M. Gabel, #2900, Parque Provincial Ernesto Tornquist, 27-1-1996, Provincia Buenos Aires; #2848, Rt. 157 south of Tucumán, 23-1-1996, Provincia Tucumán; # 2829, 3.0 km south of La Pelada, north of Santa Fe, 15-1-1996, Provincia Santa Fe.

Comments: Parbery (1967, 1971) reported this fungus on species of *Paspalum* from British Guinea and Brazil. This is the first report of the fungus in Argentina and on these host species. Although there is some variation in ascospore size among the specimens they are very similar to each other and to *P. punctum* (Parbery, 1967).

Phyllachora spartinae Orton

On *Spartina densiflora* Brongniart: Sparse mycelium forming amphigenous lesions on living leaves. Clypeus round to cylindrical, (1)-2.23-(4) X (.25)-.9-(1.5) mm, surrounded by a narrow (.1-.3 mm) chlorotic to orange halo. Perithecia not intact. Abundant, septate, filiform paraphyses 2.5 µm wide. Tips rounded. Asci cylindrical, uniseriate to partially biseriate, (85)-98-(115) X (14)-16-(20) µm, eight-spored, pedicels (5)-23-(50) µm. Ascospores hyaline, one celled, oval to broadly ellipsoid, (14)-16-(21) X (7.5)-9-(10) µm, smooth. Spermagonia and spermatia not observed. Specimens examined: A. and M. Gabel, # 2916, 10 km southwest of General Lavalle, 2-2-1996; # 2917, 23 km west of Mar de Ajó in coastal marsh, 2-2-1996, Provincia Buenos Aires.

Comments: This is a new record of the fungus in Argentina and on *S. densiflora*, which is a coastal grass. Specimens are similar to Orton's (1944) description of the fungus on *S. alterniflora* Lois. from coastal areas of North America.

Phyllachora sylvatica Sacc. & Speg.

On *Festuca purpurascens* Banks et Solander: Amphigenous, raised lesions on living leaves. Clypeus round to oval, (.25)-.52-(1) X (.25)-.4-(.75) mm, surrounded by orange halo, .25-.5 mm. Perithecia (130)-196-(239) µm wide X (96)-178-(217) µm high. Paraphyses hyaline, filiform, 2.5 µm wide. Asci cylindrical, sometimes subclavate, uniseriate to partially biseriate, (75)-83-(90) X (10)-11-(12.5) µm, eight-spored, pedicels (5)-9-(12.5) µm. Ascospores hyaline, one-celled, oval to ellipsoid, (11.3)-12.6-(13.8) X (5)-7.1-(7.5) µm, smooth. Spermagonia and spermatia not observed. Specimen examined: A. and M. Gabel, # 2876, Península Quetrihué on Rt 231, Parque Nacional Nahuei Huapí, 2-5-1996, Provincia Neuquén.

On *Festuca pampeana* Speg.: Amphigenous, raised lesions on living leaves. Clypeus oval-cylindrical, (1)-1-(1.5) X (.13)-.16-(.25) mm, surrounded by yellow-orange halo .13-.25 mm. Perithecia (217)-228-(239) µm wide X (196)-203-(218) µm high. Paraphyses hyaline, filiform, 2.5 µm wide. Asci cylindrical, uniseriate to partially biseriate, (80)-85-(105) X (10)-11.9-(15) µm, eight-spored, pedicels (15)-16.3-(17.5) µm. Ascospores hyaline, one-celled, ovoid-ellipsoid, (12.5)-14.1-(16.3) X (6.3)-7.4-(7.5) µm, smooth. Spermagonia and spermatia lacking. Specimen examined: A. and M. Gabel, # 2855, at La Pampilla, on Rt 20 west of Villa Carlos Paz and 6.0 km east of El Cóndor, 24-1-1996, Provincia Córdoba.

Comments: The fungus has been reported on *Festuca* spp. from the United States and Europe (Parbery, 1967; Orton, 1944). Dennis (1970) reported it on *Pharus* sp. from Venezuela. This is the first report of the fungus in Argentina and on *Festuca purpurascens* and *F. pampeana*.

Phyllachora vulgata Theiss. & Syd.

On *Muhlenbergia schreberi* Gmelin: Amphigenous, adaxially raised lesions on living leaves. Clypeus round to elongate, (.3)-.7-(1.3) X (.3)-.5-(.8) mm, with no halo, but with purple-brown discoloration between clypei. Perithecia (117)-127-(165) µm wide X (87)-122-(144) µm high. Abundant, septate, filiform paraphyses, 2.5 µm wide. Tips rounded. Asci cylindrical, uniseriate, (42.5)-57-(67.5) X (6)-7-(8) µm, eight-spored, pedicels (5)-7-(10) µm. Ascospores hyaline, one celled, short-oval to ovoid, (7.5)-8-(9) X (3.8)-4.6-(5) µm, smooth. Spermagonia larger than perithecia, (108)-141-(195) µm wide X (131)-158-(174) µm high. Spermatia abundant, hyaline, filiform, one-celled, (10)-12-(13) X 1.0 µm. Specimen examined: A. and M. Gabel, #2845, 4.7 km. west of La Heladera, in wet forest, 22-1-1996, Provincia Tucumán.

Comments: This is the first report of the fungus in Argentina. Parbery (1967) and Orton (1944) describe this species on several species of *Muhlenbergia* including *M. schreberi* in North America. Parbery (1967) examined a specimen on *M. sylvatica* from Uruguay.

Phyllachora **sp. 1**

On *Danthonia cirrata* Hackel et. Arechavaleta: Amphigenous, raised lesions on living leaves. Clypeus elongate to cylindrical, (1.5)-2.2-(3) X (.25)-.46-(.5) mm, surrounded by yellow halo, .25 mm wide. Perithecia sometimes at two levels, (87)-143-(261) µm wide X (87)-142-(217) µm high. Paraphyses hyaline, filiform, 1.25 µm wide. Asci cylindrical, uniseriate, (60)-69-(85) X (10)-12.6-(14) µm, eight-spored, pedicels common (2.5)-9-(15) µm. Ascospores hyaline, one-celled, oval to ovoid, (8.8)-11.6-(12.5) X (5)-7.1-(7.5) µm, smooth. Spermagonia and spermatia not observed. Specimen examined: A. and M. Gabel, # 2913, Rt. 74 east of Ayachucho, 16.5 km east of intersection with Rt 2 at Las Armas, 1-3-1996, Provincia de Buenos Aires.

Comments: Asci and ascospore size are not similar to sizes listed for *P. danthoniae* by Parbery (1967). The specimen could be a new species.

Phyllachora **sp. 2**

On *Eustachys retusa* (Lag.) Kunth (=*Chloris retusa* Lag.): Amphigenous, adaxially raised lesions on living leaves. Clypeus round to elongate, (.5)-.9-(1.5) X (.25)-.7-(1) mm, surrounded by salmon halo, changing to a pale purple near periphery. Perithecia (131)-223-(304) µm wide X (87)-151-(261) µm high. Paraphyses hyaline, filiform, 1.25 µm wide. Asci cylindrical, uniseriate, (80)-87-(95) X (7.5)-7.8-(10) µm, eight-spored, pedicels (7.5)-12.5-(22.5) µm. Ascospores hyaline, one-celled, oval to ovoid, (10)-11.1-(12.5) X (5)-6.4-(7.5) µm, pale gold, smooth. Spermagonia common, (65)-104-(131) µm wide X (96)-141-(174) µm high. Spermatia hyaline, filiform, slightly curved, (10)-11-(15) X 1.25 µm. Specimen examined: A. and M. Gabel, # 2859, Rt. 188 to Realicó at intersection with road to Secovia, 53 km south of Secovia, 29-1-1996, Provincia de San Luis.

Comments: This specimen is similar to *P. cynodontis* previously reported on *Chloris* sp. from several locations in Argentina. Ascospores are similar in size and shape and *P. cynodontis* has yellow ascospores. However, ascus width is narrower (7.8 µm) than the 12-15 µm listed by Parbery (1967). The current specimen may be immature or could be a new species.

Phyllachora **sp. 3**

On *Stipa hyalina* Nees: Amphigenous, unraised lesions on living leaves. Clypeus elongate to cylindrical, (1)-1.4-(3) X (.5)-.56-(.75) mm, with orange halo, .25-.5 mm wide. Perithecia (75)-80-(95) µm wide X (63)-67.5-(75) µm high. Paraphyses hyaline, filiform, 1.25 µm wide. Asci cylindrical, uniseriate, (52.5)-72.5-(83) X (7.5)-7.8-(10) µm, eight-spored, pedicels not observed. Ascospores hyaline, one-celled, oval to ovoid, (8.8)-10.2-(12.5) X (3.8)-4.6-(5) µm, smooth. Spermagonia uncommon, (87)-130-(196) µm wide X (65)-83-(109) µm high. Spermatia hyaline, filiform, straight to weakly curved, (11.3)-13.7-(15) X 1.25 µm. Specimen examined: A. and M. Gabel, # 2827, Rt 4, 3.0 km south of La Pelada, 15-1-1996, Provincia de Santa Fe.

Comments: Perithecia appeared immature. They could be mature perithecia of a new species or immature perithecia of an existing species.

Summary and Discussion

Forty-one specimens representing twelve species of *Phyllachora* were collected, which adds to the nineteen specimens representing thirteen species previously on record. We report seven species of *Phyllachora*, *P. chusqueae*, *P. luteo-maculata*, *P. oxyspora*, *P. punctum*, *P. spartinae*, *P. sylvatica* and *P. vulgata* not previously reported from Argentina and twenty-one species of grasses not previously listed as hosts for

Phyllachora. Three specimens of *Phyllachora* have not been identified to species and each is on a grass not previously recorded as a host. These three specimens may be new species of *Phyllachora*, but they have been listed without a species determination until more material can be examined.

Fig. 1 shows the number of specimens previously collected and provinces where they were collected, and the number of specimens collected during this study and provinces where they were collected. Collections from this study have extended the geographic distribution to include the first collections from Entre Rios, Santa Fe and Patagonia (Neuquen and Chubut).

Although the provinces where grasses were examined lie approximately between 28 to 53 degrees south latitude nearly all grasses infected with *Phyllachora* spp. were found in provinces between 27 to 39 degrees south latitude. This area includes much of the pampa and areas which are not located in the rain shadow of the Andes. The only collections from Patagonia, the southern portion of Argentina, which is comprised of Neuquen, Chubut, Santa Cruz, and Rio Negro were on *Chusquea* spp. located in *Chusquea/Nothofagus* forests on the eastern slope of the southern Andes. Most of Patagonia is in the rain shadow of the Andes and extremely arid.

Spore size, spore shape and ascus width were judged to be most stable and were the primary characters used for identification (Parbery and Langdon, 1964). The host genus of this obligate fungus (Parbery, 1967, 1971; Cannon, 1991; Orton, 1944) was an important consideration in identification. Other characters such as clypeus size and shape, arrangement of ascospores in the ascus, presence of spermagonia, presence or absence of pedicels on asci and perithecial size and shape were evaluated, but considered less important because in numerous species there was substantial variation within a specimen. Stage of development at the time of collecting would account for some of the variation. Pedicels appeared to be ephemeral, but in some specimens we observed both pedicellate and non-pedicellate mature asci. Because of this variation we listed ascus length for the sporogenous portion and pedicel length separately. Shape and size of perithecia are influenced by proximity to major vascular bundles (Gabel, 1989). We agree with Parbery and Langdon (1964) that ascus length and spore arrangement are less important taxonomic criteria because of variation within specimens.

Acknowledgments

We thank R. Hanlin (University of Georgia) and L. Tiffany (Iowa State University) for critically reading the manuscript. L. Clark (Iowa State University) graciously identified the species of *Chusquea*. The project was partially funded by a Faculty Research Grant from Black Hills State University.

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MYCOTAXON

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FUNGI ON JUNCUS ROEMERIANUS.
13. HYPHOPOLYNEMA JUNCATILE SP. NOV.

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ABSTRACT

The inconspicuous sporodochia of *Hyphopolynema juncatile* sp. nov. develop on senescent standing leaves of *Juncus roemerianus* in salt marshes on the coast of North Carolina. The new species occurs between the lower and upper parts of the leaves and is considered to be a facultative marine fungus. It is compared with the other four species of *Hyphopolynema*.

INTRODUCTION

In the continuing survey of the mycota of the halophyte *Juncus roemerianus* Scheele, so far we have described 32 ascomycetes (Kohlmeyer *et al.* 1999 and preceding articles of this series). Additionally, we proposed one member of Agonomycetales (Kohlmeyer & Volkmann-Kohlmeyer 1996a), one coelomycete (Kohlmeyer & Volkmann-Kohlmeyer 1996b) and three hyphomycetous states of ascomycetes, viz., *Trichocladium medullare* Kohlm. & Volkm.-Kohlm. (anamorph of *Gaeumannomyces medullaris* Kohlm. *et al.*), *Cirrenalia adarca* Kohlm. *et al.* (anamorph of *Juncigena adarca* Kohlm. *et al.*) and *Dwayaangam junci* (anamorph of *Orbilia junci* Kohlm. *et al.*) (Kohlmeyer *et al.* 1995, 1997, 1998, Kohlmeyer & Volkmann-Kohlmeyer 1995). In the following we describe a new hyphomycete in the genus *Hyphopolynema* Nag Raj.

Hyphopolynema juncatile Kohlm. & Volkm.-Kohlm., anam.-sp. nov.
Etymology: From the host genus and the Latin suffix *-atilis*, indicating the place of growth.

Conidiomata *150-220 μm diam.,* 25-35 *μm alta, sporodochialia,*

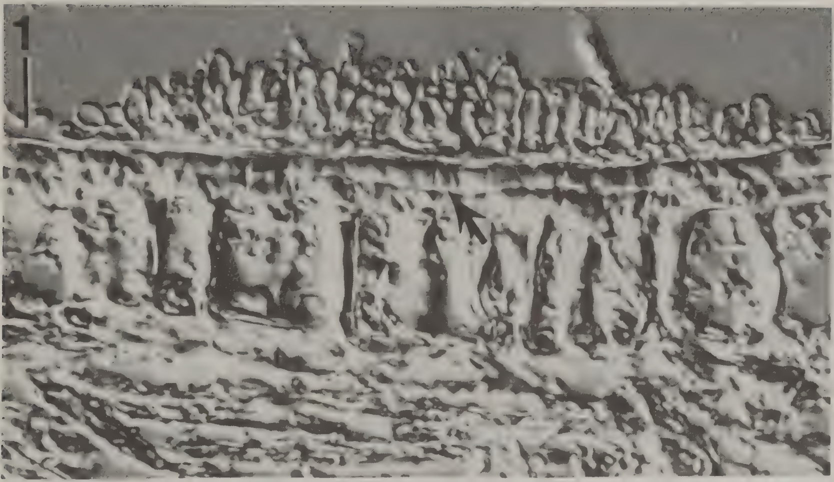
circularia, superficialia, facile excussa ab epidermide, cum vel sine setis, singularia, brunnea ubi exsiccata, dilute aurantiaca ubi humida. Setae *sparsae, periphericae, subulatae, rectae, altro-brunneae, 3-5-septatae, in apicem gradatim decrescentes, 55-90 μm longae, 4-7 μm diam. ad basem, 2.5-3.5 ad apicem.* Conidiophora *ad cellulas conidiogenas reducta, cylindrica, monophialidica, dilute brunnea, hymenium compactum formantia.* Conidia *13-16 x 3-4 μm ($\bar{x}$ = 14.2 x 3.6 μm; n = 50), fusiformia ad ellipsoidea, uniseptata in medio, ad septum non constricta, base rotundata, hyalina; appendice singulare apicale et 3-4 appendicibus basalibus; appendices 7-10 μm longae, filamentosae.*

SUBSTRATUM: *Folia Junci roemeriani.*
DISTRIBUTIO: *Littus Oceani Atlantici, America Septentrionalis* (U.S.A.: North Carolina).
HOLOTYPUS: J.K. 5614 **(IMS)**; ISOTYPUS: J.K. 5614 **(NY).**

Conidiomata 150-220 μm diam, 25-35 μm high (external part), sporodochial, circular in outline, forming a superficial flat plate (Figs. 1-4, 7), penetrating the cuticle and epidermis with thin hyphae (Figs. 1,2,4,6), proliferating intracellularly in the cortical tissue of the host and here forming a brown pseudostroma with *textura angularis* (Fig. 4); external part easily dislodged from the substrate (Fig. 5), with or without setae, single; brown when dry, on incubated leaves in a moist chamber appearing pale orange. *Setae* sparse, peripheral, subulate, erect, straight, 3-5-septate, tapered towards the apex, 55-90 μm long, 4-7 μm in diam. at the dark brown base, (2.5-3.5 μm at the light brown tip. *Conidiophores* reduced to conidiogenous cells; these are cylindrical, monophialidic, light brown, and arranged in a compact hymenium, forming a *textura porrecta* in longitudinal section (Figs. 2-5), originating from the polygonal upper cells of the conidioma. *Conidia* 13-16 x 3-4 μm ($\bar{x}$ = 14.2 x 3.6 μm; n = 50), holoblastic, fusiform to ellipsoidal, one-septate in the middle, not constricted at the septum, hyaline with a rounded base, with one apical and 3-4 basal filamentous appendages, 7-10 μm long (Figs. 5,7,8).

SUBSTRATUM: Senescent leaves of *J. roemerianus.*
DISTRIBUTION: Atlantic Coast (U.S.A.: North Carolina).
MATERIAL EXAMINED: Dead standing leaves of *J. roemerianus,*

Figs. 1-3. *Hyphopolynema juncatile.* 1 and 2. Young sporodochia, closely attached to the cuticle and penetrating it (arrows), 4 μm longitudinal sections. 3. Mature sporodochium, hyphal masses in the epidermal cells (arrow), 6 μm longitudinal section. Figs. 1 and 2 from J. K. 5614, Fig. 3 from J. K. 5615. Scale bars: Fig. 1, 10 μm, Figs. 2 and 3, 20 μm.



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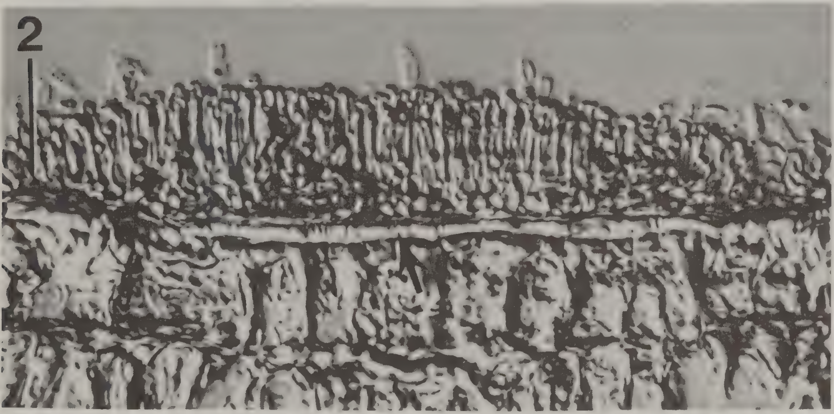

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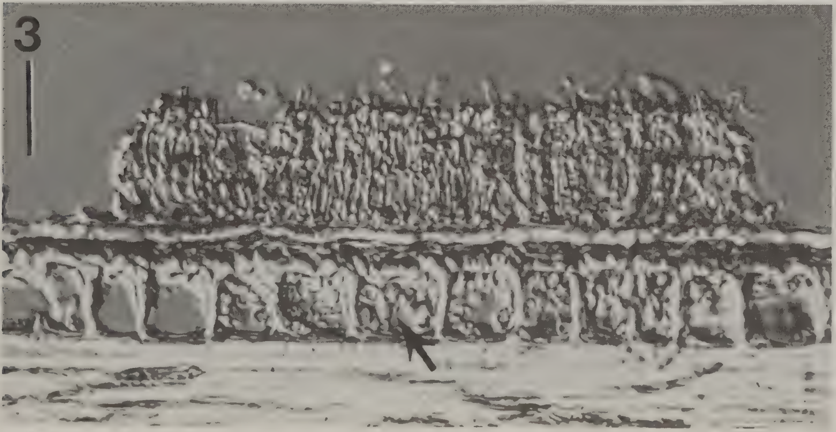

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Broad Creek, Carteret County, North Carolina (34°43'N, 76°55'07"W): 12 Oct. 1997, J. K. 5614 (Holotype: **IMS**; Isotype: **NY**; also in pure culture); same location: 23 Apr. 1995 (J. K. 5554), 13 May 1995 (J. K. 5556), 5 Mar. 1997 (J. K. 5607), 30 Oct. 1997 (J. K. 5615).

We found sporodochia of *H. juncatile* mostly in the tips of senescent leaves, but on one occasion as low as 33 cm above the rhizome. Therefore, the fungus can be classified as a facultative marine species. The flat sporodochia are inconspicuous and difficult to detect on dry leaves. This may explain the relative small number of collections. Most probably the fungus occurs throughout the year.

We recognize the circumscription of *Hyphopolynema* as emended by Sutton & Alcorn (1984) to include setose species and place the new species from *Juncus roemerianus* in this genus. The fact that setae are rare in *H. juncatile* and may be absent in some or most sporodochia further supports the view that the presence or absence of setae is unimportant at the generic level. So far, four species of *Hyphopolynema* have been described. The type species, *H. tropicale* Nag Raj, occurs on pods of *Inga spectabilis* (Fabaceae) and is distinguished from *H. juncatile* by mostly one-celled conidia up to 19 μm long and 5 μm wide (up to 7.5 μm wide, according to Sutton & Alcorn 1984), with a truncate base (Nag Raj 1977). The Australian species, *H. australe* B. Sutton & Alcorn and *H. ellisiorum* B. Sutton & Alcorn (Sutton & Alcorn 1984) are also distinctly different from *H. juncatile.* The former grows on the pteridophyte *Blechnum indicum*, has dark brown setae scattered throughout the conidiomata and thin, 2-2.5 μm wide conidia. The latter develops on decomposing *Juncus effusus,* has peripheral or sometimes scattered setae and conidia with a truncate base, forming straw-colored or pale luteous masses. Finally, *H. stilboideum* Bhat & W. B. Kendr. has synnematal conidiomata growing on dead leaves of *Syzygium* (Myrtaceae) in India (Bhat & Kendrick 1993).

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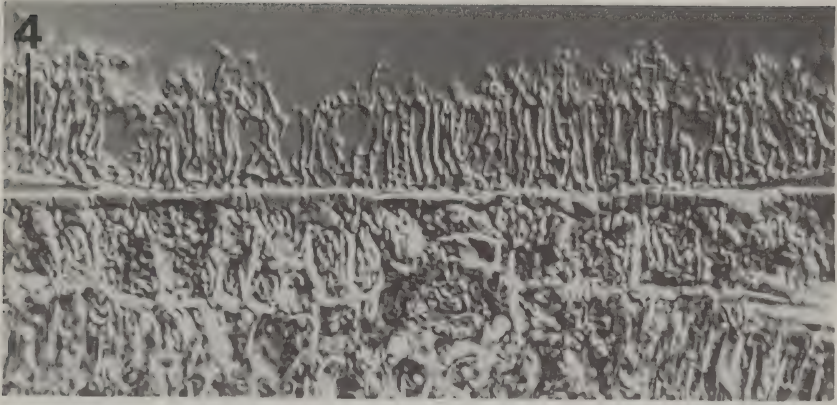
We are grateful to Dr. R. D. Goos for serving as a presubmission reviewer. We thank L. White and H. Barnes for assistance with the preparation of the manuscript.

Figs. 4-6. *Hyphopolynema juncatile*. 4. Sporodochium on the host surface with intracellular proliferation of hyphae and formation of a pseudostroma in the outer tissues, hand section. 5. Sporodochium, detached from the cuticle showing origin of conidia, hand section. 6. Surface view of epidermal cells after removal of sporodochium, pitted by penetrating hyphae. Figs. 4 and 5 from J. K. 5556. Fig. 6 from J. K. 5614. Scale bars: Figs. 4 and 5, 20 μm, Fig. 6, 10 μm.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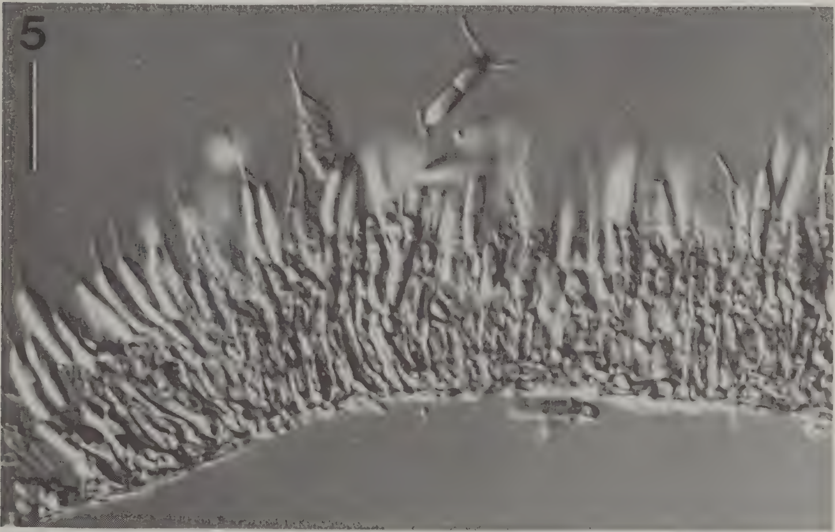
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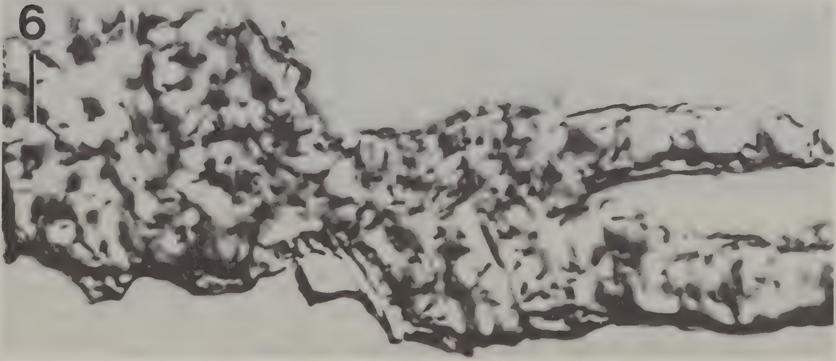
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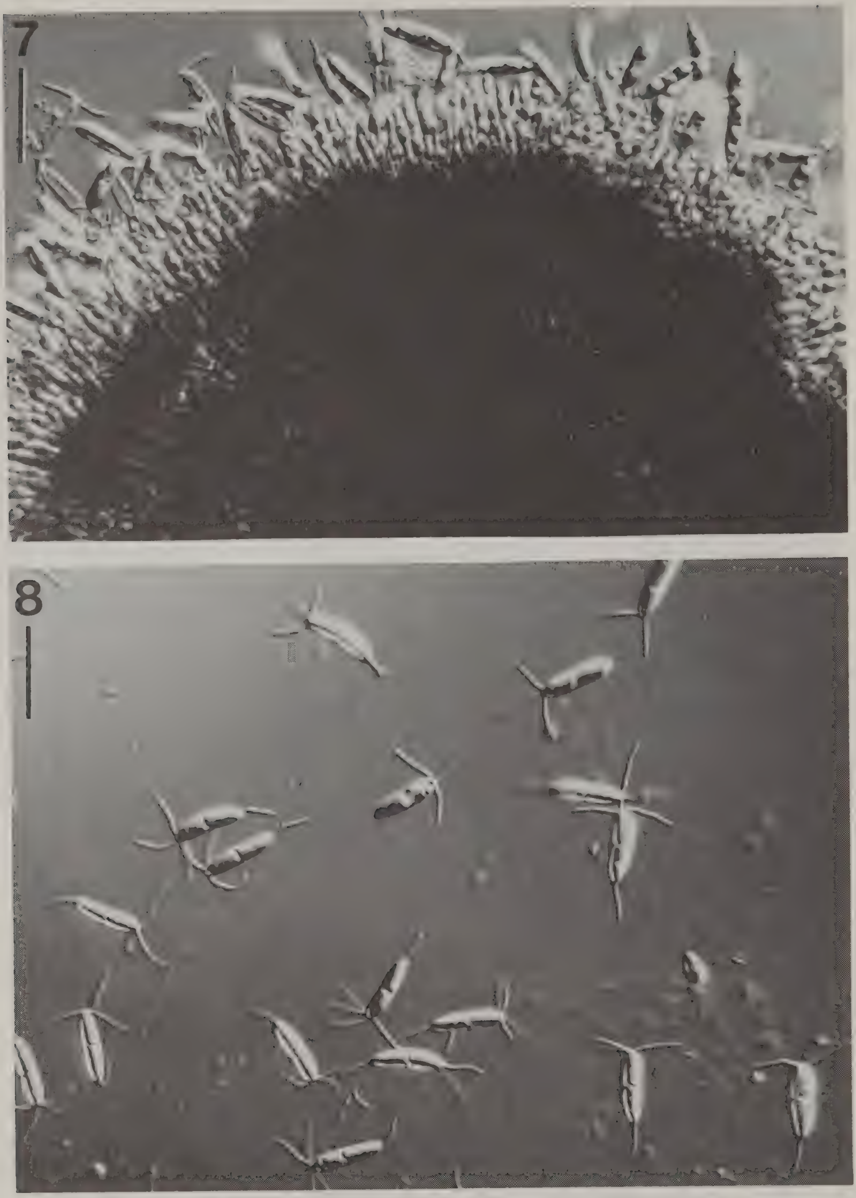

Figs. 7 and 8. *Hyphopolynema juncatile*. 7. View on top of sporodochium, showing arrangement of conidia. 8. Conidia. Fig. 7 from J. K. 5615, Fig. 8 from 5556. Scale bars: 20 μm.

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ONLINE RESOURCES FOR FUNGAL TAXONOMY AND NOMENCLATURE:

Internet Monography

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htpp://biodiversity.uno.edu/~fungi/

The internet is a medium of such marvelously protean and vibrant qualities that any attempt to pin it down in such an enduring medium as print seems the height of folly. Nevertheless, I intend in this column to present a guide to useful resources, and to highlight how the extraordinary potential of the internet can be realized.

A most exciting property of the internet is the unprecedented access to information that it can provide. We are just beginning to explore its potential in serving resources that include collection catalogs, glossaries, phone books, photographs, and books. It is marvelous venue for the publication of taxonomic monographs, as it effectively bypasses the problem of production costs that are high relative to narrow market appeal, and allows broad access to taxonomic resources.

As yet, few internet sites present taxonomic monographs on fungi. A few examples are discussed below. Granting agencies increasingly expect online dissemination of research results; for example, the US National Science Foundation's PEET competitions (Partnership for Enhancing Expertise in Taxonomy) stipulate that funded taxonomic projects will be made available online.

NSF PEET homepage	http://www.nhm.ukans.edu/~peet/

Existing online monographs are mostly adaptations of previously published works: Malloch's guide to *Moulds* and Kendrick's textbook, *The Fifth Kingdom*. The scanned text of these works has been enhanced with color illustrations, animated image files, cross-indexing and links to related sites, and both make remarkable teaching tools.

MOULDS: ISOLATION, CULTIVATION, IDENTIFICATION
http://www.botany.utoronto.ca/ResearchLabs/MallochLab/Malloch/Moulds/Contents.html
THE FIFTH KINGDOM http://www.pacificcoast.net/~mycolog/fifthtoc.html

Beyond the basics, internet sites can take advantage of the special capabilities of the internet to enhance content. Three particularly good exemplars of monographic web sites are one on the Xylariaceae by Rogers, Ju and Adams; Mueller's monograph of North American *Laccaria* species; and Halling's monograph of North American *Collybia* species. These too have been adapted from published manuscripts with thoughtful expansion.

HOME OF THE XYLARIACEAE	http://mycology.wsu.edu/Xyalariaceae/
THE MUSHROOM GENUS *LACCARIA*	http://www.fmnh.org/candr/botany/botany_sites/fungi/index.html
A REVISION OF *COLLYBIA* S. L. IN THE NORTHEASTERN UNITED STATES AND ADJACENT CANADA	htpp://www.nybg.org/bsci/res/col/colintro.html

What makes a user-friendly monograph? The same things that define a good printed monograph: careful taxonomy and detailed description, thorough keys, high quality illustrations, good indices. The internet-savvy monographs listed above include menu bars that facilitate navigation, extensive cross-indexing (hyperlinking), many high quality images, and thoughtfully designed formats (each slightly different). Synoptic keys come into their own on the WWW (more of these in a future column): FusKey and the Genus Finder of PezWeb are excellent examples.

PEZWEB	http://www/bcc.orst.edu/mycology/pezweb/
FUSKEY	http://res.agr.ca/brd/fusarium/

Illustration can be provided in many formats outside the constraints of expensive paper publication, including high and low resolution versions and animated images. Online monographs might include links to culture collections and herbarium databases, and even provide maps of species distribution that are customizable in scale and in the superposition of human and environmental features. The can be run from master databases, facilitating future updates and (carefully documented) error correction.

Perhaps the greatest potential problem of online monography is that of information quality: peer review has traditionally ensured the reliability of published works, but it remains to be seen what review system may develop online. The validity and import of internet publications during tenure review and performance appraisal is not yet firmly established, due in part to the common lack of peer review. New species names cannot validly be published on the internet under the current International Code of Botanical Nomenclature. And all website developers must take into account that access to the internet is not universally available or inexpensive, and that complicated pages may be frustratingly slow to download.

Five years ago I could find only sixteen mycological resources on the internet; today my website, *Mycological Resources on the Internet,* lists over four hundred, an indication of the dynamism of the internet. Despite its problems, the fluid qualities of the internet promise to revolutionize access to taxonomic resources.

All websites reviewed in this column are accessible through:

Mycological Resources on the Internet	http://biodiversity.uno.edu/~fungi/.

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BOOK REVIEWS

Eric H.C. McKenzie, Book Review Editor

A Nomenclatural Study of the Ganodermataceae Donk by J.-M. Moncalvo and L. Ryvarden (1997). 114 pp., 220 x 155 mm. *Synopsis fungorum 11.* Available from Fungiflora A/S, P.O. Box 95, Blindern, N-0314, Oslo, Norway (e-mail: jeanmarc@duke.edu). ISBN 82-90724-18-7. NOK180 + postage.

The need for this publication is implicit in the proposal of Ryvarden (1995): "That no new species be described in *Ganoderma* in the decade to 2005 and those authors breaking this law be sentenced to do taxonomic work on at least 10 old names"! This nomenclatural treatment of the family Ganodermataceae, while giving offending authors a helping hand, provides a foundation for meaningful advances in taxonomy of these fungi. With many species described from one or few collections and based on narrow species concepts, circumscription of taxa in Ganodermataceae is poor and there is urgent need to compare species, stabilise names, and differentiate members of species complexes. The family includes several species of importance to plant pathology and medicine, the names of which remain uncertain. Both authors have published extensively on *Ganoderma* and related genera, and combine molecular expertise with morphological research on field-collected and herbarium material.

The book provides the first modern compilation of names of species in the Ganodermataceae (368 in total), including synonymy and nomenclatural and taxonomic status of each name. *Ganoderma*, as one of the largest genera of polypore fungi, contains 148 of the 220 "species" recognised in the family. Treatment of names is based on an extensive review of literature, and does not typically include new taxonomic evaluation except for assignment of each species to a taxonomic group.

The past 200 years of taxonomic history of Ganodermataceae is reviewed (Section 2), detailing the varying treatments of genera and taxa at subgeneric ranks and reflecting changing emphasis from macro-morphology to micro-morphology, and more recently to cultural and molecular studies. Phylogeny is discussed briefly, with morphological and molecular studies supporting recent divergence of *Ganoderma*. The authors then list (Section 4) all names used at each taxonomic rank from Order to Section, accompanied by a statement on status. Descriptions of genera are not provided; instead the authors briefly define six morphological "*genus* groups": *Ganoderma*, *Elfvingia*, *Amauroderma*, *Tomophagus*, *Humphreya*, and *Haddowia*. In so doing, there is apparently no intention to accept *Elfvingia* at generic rank. Subgenera and sections, while used extensively for *Ganoderma* by some authors (e.g., Steyaert), are not discussed in detail and are not utilised in the species listings.

The list of species names (Section 6) occupies most of the book, with names arranged alphabetically by epithet. Note that the genus name follows the authority and is reduced to the first letter(s) only; it may sometimes be mistaken for an author's initial since unlike the epithet it is not in bold type. A few typographical errors were also noted in this section.

For each species name, full citation of authority, reference, and type specimen, including herbarium location, is provided followed by synonyms. For taxonomic synonyms, reference is cited to the author(s) who proposed each synonymy with a useful indication of whether the synonymy is supported by examination of type specimens (as "teste author"). The nomenclatural status of each name is given. Although new synonymies and new combinations have not been proposed, it is worth noting validation of *G. subamboinense* (Henn.) Bazzalo & Wright ex Moncalvo & Ryvarden.

Following each accepted species name, the appropriate taxonomic group is designated, and references made to published descriptions and taxonomic comment. For confused and inadequately typified names, the authors synthesise published treatments and recommend that names be reassessed, abandoned, or neotypified. The geographic distribution of each species is also given.

Many species are considered to belong to one of six or more complexes (e.g., *G. applanatum - australe* complex, *G. lucidum* complex) although these complexes are not discussed in detail. A list of species belonging to each complex would be useful; this, in part, is provided by the arrangement of species in the distribution tables in Section 7, although the only complex named there is *G. chalceum - boninense* complex, elsewhere referred to as *G. chalceum* complex.

In a series of tables in Section 7, species are grouped according to their distribution by broad geographical region. This summary will greatly assist rationalisation of superfluous names. The tables also highlight the frequency of taxa known from only the type locality or adjacent areas. The Pacific region is somewhat ambiguously defined, and by deduction should include New Zealand and Australia. The geographic listings are also used to indicate synonymy and status of typification of names, designated by defined symbols. Species names are at times difficult to locate in these tables, however, since within their relevant "genus group" species are arranged under their main region of distribution, rather than alphabetically. Distribution data is summarised in Conclusions (Section 8); the reported absence of *Amauroderma* from temperate regions appears to overlook Cunningham's (1965) record of *A. rude* from Tasmania and southern Australia.

The authors have prepared a valuable and exhaustive synthesis of published treatments of all species of Ganodermataceae, and have assessed nomenclatural and taxonomic status of many confused taxa. Considerable work remains to develop a stable taxonomy for these fungi, but this publication will both stimulate and form the basis for all future taxonomic studies of the family.

References

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Ryvarden, L. 1995: Can we trust morphology in *Ganoderma*? In: Buchanan, P.K.; Hseu, R.S.; Moncalvo, J.M. (ed.). *Ganoderma*, Systematics, Phytopathology and Pharmacology.

Proceedings of Contributed Symposium 59 A,B, 5th International Mycological Congress, Vancouver, August 14-21, 1994. Taipei. p. 19-24.

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Cercosporoid Fungi from Russia and Adjacent Countries by U. Braun and V.A. Melnik (1997). 112 pp., 84 figures, 1 map, 290 x 205 mm. *Russian Academy of Sciences, Proceedings of the Komarov Botanical Institute, Volume 20.* Available from Koeltz Scientific Books, P.O. Box 1360, D-61453 Koenigstein, Germany (e-mail: koeltz@ibm.net), or Koeltz Scientific Books, 1911 North Duncan Road, Champaign, Illinois 61821, U.S.A. (e-mail: koeltz@shout.net). ISBN 5-201-11094-0. Approx. US$20.

This publication is primarily an alphabetical index to all those species of cercosporoid fungi known from the former USSR, either through the literature, or from specimens deposited in herbaria LE or LEP. It provides up-to-date names, where necessary cross referenced from unaccepted names, and is the first comprehensive treatment of these fungi for this part of the world since 1937. The fungal names are listed together with synonyms, literature references, hosts and distribution within the defined geographic area. For many species there are notes giving details of specimens examined, or in some cases indicating the unavailability of specimens, and there are often short morphological descriptions. Many specimens have been examined, including all types described from the former USSR area, and other critical species. There are 84 line drawings illustrating conidiophores and conidia of selected species. For those with poor geographic knowledge there is a fold out map of the area covered, and 50 regions within Russia are named.

Introductory sections provide a brief history of the study of cercosporoid fungi in the region, a generic description of the 16 genera which are covered, and a list of host genera with names of cercosporoid fungi recorded on them - accepted fungal species are in bold. There are also two keys to the genera - a dichotomous key, and a synoptic key. The latter includes only the 16 treated genera (although *Distocercospora* is in the key, but it appears not to occur in the area), while the dichotomous key includes cercosporoid genera which do not occur in the area, as well as some allied genera with which they may be confused.

Many taxonomic changes are made in this book including the description of a new genus (*Pseudophaeoramularia*), four new species, and one new variety, and 43 new combinations are proposed. Some other species are reduced to synonymy. The taxonomic changes do not necessarily refer only to Russian fungi, e.g., the new combinations *Passalora chamaecristae* and *P. gerardiae* refer to species from North America.

In recent years *Cercospora* sensu lato has been split into numerous smaller genera, several of which have been introduced by Braun. It is, perhaps, misleading to state that these smaller genera are "more natural units", especially as the teleomorphs for all these fungi so far belong in *Mycosphaerella*.

It is interesting to note that *Cercospora physalidis* is treated as the *C. apii*-like counterpart on hosts belonging to the Solanaceae. Thus, well-known species such as *C. capsici*, *C.*

melongenae, C. nicotianae, and *C. solanicola* on important economic hosts are all regarded as synonyms of *C. physalidis.* The authors suggest, however, that comprehensive cross inoculation experiments are necessary to substantiate the cited synonymy.

This publication is clearly laid-out, but there is a peculiar mixture of paragraphs which are fully justified and others which have some slightly short lines. There are a few minor typographical errors but these do not detract from the overall presentation. There is a wealth of detail on fungi from a region of the world from which such information, in English, is usually inaccessible. Many of the species treated occur in other parts of the world, and hence this publication contains information which is applicable not only to the area covered by the former USSR. The book is low-priced, but be quick, it was apparently produced in very limited numbers!

E. McK.

Mycenae Paranaenses by R.A. Maas Geesteranus and A.A.R. de Meijer (1997). 164 pp., 240 x 170 mm. *Koninklijke Nederlandse Akademie van Wetenschappen Verhandelingen, Afd. Natuurkunde, Tweede Reeks, deel 97.* Available from Royal Netherlands Academy of Arts and Sciences, P.O. Box 19121, 1000 GC Amsterdam, the Netherlands. North-Holland, Amsterdam/Oxford/New York/Tokyo. ISBN 0-444-85817-2. Dfl.125.

The state of Paraná is found in southern Brazil and "consists of a narrow coastal zone, separated by a mountain range called Serra de Mar". In general, it is a wet subtropical climate with hot summers, rare frosts, summer rainfall, and the absence of a dry season with 12-40 cm of annual precipitation. Most of the collecting sites which form the basis for this monograph are in the vicinity of Curitiba, the capital of Paraná.

A total of 63 species of *Mycena* are included, with 56 newly described taxa. A key to the sections of the genus is presented at the beginning and then individual sectional keys to species are located in the text with each section. Both macro and microscopic descriptions are provided, along with drawings of the fruiting body and good drawings of critical microscopic features such as spores and cystidia. A most valuable part of the book is a ten page key to the "South American species of *Mycena* described (or mentioned) by Singer", presented at the end of the monograph. Each species in the key is referenced to the literature which is of great value to the user. No descriptions are given but the references listed can be easily obtained for the 182 entries in the key. Some phenotypically variable species are keyed out more than once. The vast majority of the species were described by Singer, and are found from Costa Rica to South America

This monograph both reports on, and makes available, a wealth of information on taxa in the genus *Mycena* in Central and South America. The book has a substantial hard cover, it is nicely produced, and is a valuable addition to our knowledge of the Agaricales, and species in the genus *Mycena*, in particular.

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